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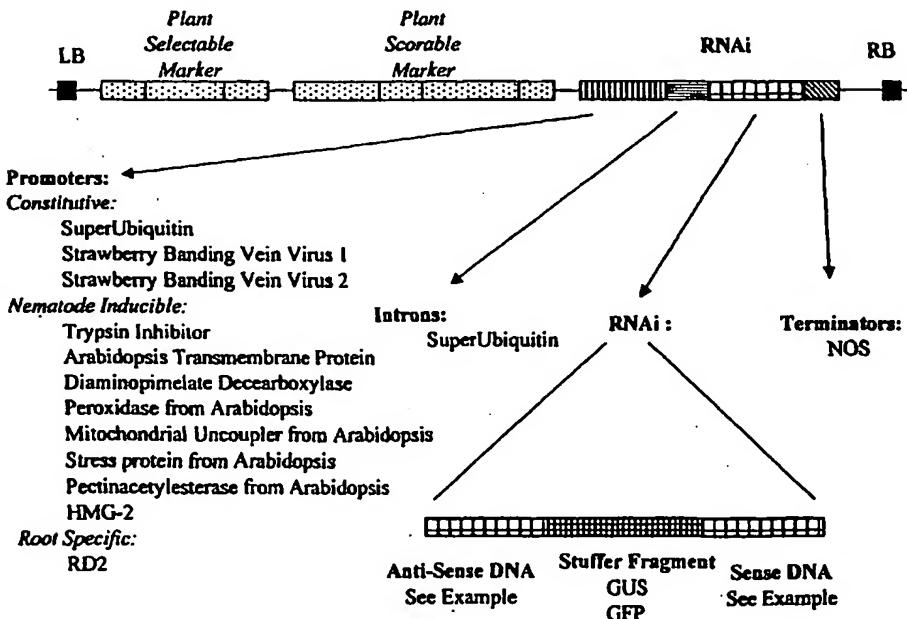
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[Continued on next page]

(54) Title: MATERIALS AND METHODS FOR THE CONTROL OF NEMATODES



(57) Abstract: The subject invention provides novel methods and compositions for controlling nematodes. More specifically, the subject invention provides RNAi molecules, polynucleotide sequences, and methods of using these sequences in nematode control.

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DESCRIPTIONMATERIALS AND METHODS FOR THE CONTROL OF NEMATODESBackground of the Invention

[0001] Plant parasitic nematodes, such as root-knot nematodes (*Meloidogyne* species) and cyst nematodes (*Globodera* and *Heterodera*), attack nearly every food crop, and are among the world's most damaging agricultural pests. For example, root-knot nematodes parasitize more than 2,000 plant species from diverse plant families and represent a tremendous threat to crop production world-wide. These biotrophic pathogens have evolved highly specialized and complex feeding relationships with their hosts.

[0002] Nematodes cause millions of dollars of damage each year to turf grasses, ornamental plants, and food crops. Efforts to eliminate or minimize damage caused by nematodes in agricultural settings have typically involved the use of soil fumigation with materials such as chloropicrin, methyl bromide, and dazomet, which volatilize to spread the active ingredient throughout the soil. Such fumigation materials can be highly toxic and may create an environmental hazard. Various non-fumigant chemicals have also been used, but these too create serious environmental problems and can be highly toxic to humans.

[0003] Some research articles have been published concerning the effects of δ -endotoxins from *B. thuringiensis* species on the viability of nematodes. See, for example, Bottjer, Bone and Gill ([1985] *Experimental Parasitology* 60:239-244); Ignoffo and Dropkin (Ignoffo, C.M., Dropkin, V.H. [1977] *J. Kans. Entomol. Soc.* 50:394-398); and Ciordia, H. and W.E. Bizzell ([1961] *Jour. of Parasitology* 47:41 [abstract]). Several patents have issued describing the control of nematodes with *B.t.* See, for example, U.S. Patent Nos. 4,948,734; 5,093,120; 5,281,530; 5,426,049; 5,439,881; 5,236,843; 5,322,932; 5,151,363; 5,270,448; 5,350,577; 5,667,993; and 5,670,365. The development of resistance by insects to *B.t.* toxins is one obstacle to the successful use of such toxins.

[0004] The pesticidal activity of avermectins is well known. The avermectins are disaccharide derivatives of pentacyclic, 16-membered lactones. They can be divided into four major compounds: A_{1a}, A_{2a}, B_{1a}, and B_{2a}; and four minor compounds: A_{1b}, A_{2b}, B_{1b}, and B_{2b}. The isolation and purification of these compounds is also described in U.S. Patent No. 4,310,519, issued January 12, 1982. Avermectin B_{2a} is active against the root-knot nematode, *Meloidogyne incognita*. It is reported to be 10-30 times as potent as commercial contact nematicides when incorporated into soil at 0.16-0.25 kg/ha (Boyce Thompson Institute for Plant Research 58th Annual Report [1981]; Putter, I. *et al.* [1981] "Avermectins: Novel Insecticides, Acaracides, and Nematicides from a Soil Microorganism," *Experientia* 37:963-964). Avermectin B_{2a} is not toxic to tomatoes or cucumbers at rates of up to 10 kg/ha.

[0005] Fatty acids are a class of natural compounds which occur abundantly in nature and which have interesting and valuable biological activities. Tarjan and Cheo (Tarjan, A.C., P.C. Cheo [1956] "Nematocidal Value of Some Fatty Acids," Bulletin 332, Contribution 884, Agricultural Experiment Station, University of Rhode Island, Kingston, 41 pp.) report the activity of certain fatty acids against nematodes. In 1977 Sitaramaiah and Singh (Sitaramaiah, K., R.S. Singh [1977] *Indian J. Nematol.* 7:58-65) also examined the response of nematodes to fatty acids. The results of these tests with short chain acids were equivocal, showing nematode-inhibitory action in some instances and stimulatory activity in other instances. Phytotoxicity of these acids was observed at higher concentrations. The short chain fatty acids were also examined by Malik and Jairajpuri (Malik, Z., M.S. Jairajpuri [1977] *Nematol. medit.* 12:73-79), who observed nematode toxicity at high concentrations of the fatty acids.

[0006] Notwithstanding the foregoing (some of the limitations of and problems associated with these approaches are discussed above), there is a need for safe and effective alternatives for controlling nematodes.

[0007] One method for disrupting normal cellular processes is by the use double-stranded interfering RNA (RNAi), or RNA-mediated interference (RNAi). When RNAi corresponding to a sense and antisense sequence of a target mRNA is introduced into a cell, the targeted mRNA is degraded and protein translation of that message is stopped. Although not yet fully understood, the mechanism of this post-transcriptional gene

silencing appears to be at least partially due to the generation of small RNA molecules, about 21 - 25 nucleotides in length, that correspond to the sense and antisense pieces of the RNAi introduced into the cell (Bass, B. L. [2000] "Double-stranded RNA as a template for gene silencing" *Cell* 101:235-238).

[0008] The specificity of this gene silencing mechanism appears to be extremely high, blocking expression only of targeted genes, while leaving other genes unaffected. A recent example of the use of RNAi; to inhibit genetic function in plants used *Agrobacterium tumefaciens*-mediated transformation of *Arabidopsis thaliana* (Chuang, C.-F. and E. M. Meyerowitz [2000] "Specific and heritable genetic interference by double-stranded RNA in *Arabidopsis thaliana*" *Proc. Natl. Acad. Sci. USA* 97:4985-4990). Chuang *et al.* describe the construction of vectors delivering variable levels of RNAi targeted to each of four genes involved in floral development. Severity of abnormal flower development varied between transgenic lines. For one of the genes, AGAMOUS (AG), a strong correlation existed between declining accumulation of mRNA and increasingly severe phenotypes, suggesting that AG-specific endogenous mRNA is the target of RNAi.

Brief Summary of the Invention

[0009] The subject invention provides novel methods and compositions for controlling nematodes. More specifically, the subject invention provides polynucleotide sequences that encode nematode genes, RNAi that selectively targets mRNA transcripts of these essential nematode genes, and methods of using these sequences in nematode control strategies. Such sequences for use according to the subject invention are summarized in Appendix 1. RNAi molecules disclosed herein can be used to inhibit the expression of one or more of these genes in nematodes.

Brief Description of the Drawings

[00010] **Figure 1:** Modular Binary Construct System (MBCS): A series of six, 8-base cutter restriction enzyme sites has been placed between the left and right Ti borders of a previously created kan^R/tet^R binary plasmid.

[00011] **Figure 2:** An exemplary shuttle vector created for cloning of useful DNA fragments by containing the multi-cloning site (MCS) of a modified Bluescript plasmid flanked by 8-base restriction sites.

[00012] **Figure 3:** An exemplary shuttle vector with exemplary inserts.

[00013] **Figure 4:** A suggested RNAi binary vector with exemplary inserts.

[00014] **Figure 5:** Exemplary selectable markers for MBCS.

[00015] **Figure 6:** Exemplary scorable markers for MCBS.

[00016] **Figure 7:** Exemplary RNAi binary vector.

[00017] **Figure 8:** Exemplary RNAi shuttle vector.

Brief Description of the Sequences

[00018] Brief Description of the Sequences can be found in Appendix I.

Detailed Disclosure of the Invention

[00019] The subject invention provides novel methods and compositions for controlling nematodes. More specifically, the subject invention provides polynucleotide sequences and methods of using these sequences in nematode control strategies. A preferred method for controlling nematodes according to the subject invention provides materials and methods for controlling nematodes by using double-stranded interfering RNA (RNAi), or RNA-mediated interference (RNAi). The terms RNAi and RNAi are used interchangeably herein unless otherwise noted.

[00020] In one embodiment of the invention, RNAi molecules are provided which are useful in methods of killing nematodes and/or inhibiting their growth, development, parasitism or reproduction. RNAi molecules of the invention are also useful for the regulation of levels of specific mRNA in nematodes.

[00021] dsRNA (RNAi) typically comprises a polynucleotide sequence identical to a target gene (or fragment thereof) linked directly, or indirectly, to a polynucleotide

sequence complementary to the sequence of the target gene (or fragment thereof). The dsRNA may comprise a polynucleotide linker (stuffer) sequence of sufficient length to allow for the two polynucleotide sequences to fold over and hybridize to each other; however, a linker sequence is not necessary. The linker (stuffer) sequence is designed to separate the antisense and sense strands of RNAi significantly enough to limit the effects of steric hindrances and allow for the formation of dsRNA molecules.

[00022] RNA containing a nucleotide sequence identical to a fragment of the target gene is preferred for inhibition; however, RNA sequences with insertions, deletions, and point mutations relative to the target sequence can also be used for inhibition. Sequence identity may be optimized by sequence comparison and alignment algorithms known in the art (see Gribskov and Devereux, *Sequence Analysis Primer*, Stockton Press, 1991, and references cited therein) and calculating the percent difference between the nucleotide sequences by, for example, the Smith-Waterman algorithm as implemented in the BESTFIT software program using default parameters (e.g., University of Wisconsin Genetic Computing Group). Alternatively, the duplex region of the RNA may be defined functionally as a nucleotide sequence that is capable of hybridizing with a fragment of the target gene transcript.

[00023] As disclosed herein, 100% sequence identity between the RNA and the target gene is not required to practice the present invention. Thus the invention has the advantage of being able to tolerate sequence variations that might be expected due to genetic mutation, strain polymorphism, or evolutionary divergence.

[00024] RNA may be synthesized either *in vivo* or *in vitro*. Endogenous RNA polymerase of the cell may mediate transcription *in vivo*, or cloned RNA polymerase can be used for transcription *in vivo* or *in vitro*. For transcription from a transgene *in vivo* or an expression construct, a regulatory region (e.g., promoter, enhancer, silencer, splice donor and acceptor, polyadenylation) may be used to transcribe the RNA strand (or strands). Inhibition may be targeted by specific transcription in an organ, tissue, or cell type; stimulation of an environmental condition (e.g., infection, stress, temperature, chemical inducers); and/or engineering transcription at a developmental stage or age. The RNA strands may or may not be polyadenylated; the RNA strands may or may not be capable of being translated into a polypeptide by a cell's translational apparatus. RNA

may be chemically or enzymatically synthesized by manual or automated reactions. The RNA may be synthesized by a cellular RNA polymerase or a bacteriophage RNA polymerase (e.g., T3, T7, SP6). The use and production of an expression construct are known in the art (see, for example, WO 97/32016; U.S. Pat. Nos. 5,593,874; 5,698,425; 5,712,135; 5,789,214; and 5,804,693; and the references cited therein). If synthesized chemically or by *in vitro* enzymatic synthesis, the RNA may be purified prior to introduction into the cell. For example, RNA can be purified from a mixture by extraction with a solvent or resin, precipitation, electrophoresis, chromatography, or a combination thereof. Alternatively, the RNA may be used with no or a minimum of purification to avoid losses due to sample processing. The RNA may be dried for storage or dissolved in an aqueous solution. The solution may contain buffers or salts to promote annealing, and/or stabilization of the duplex strands.

[00025] Preferably and most conveniently, RNAi can be targeted to an entire polynucleotide sequence of a gene set forth herein. Preferred RNAi molecules of the instant invention are highly homologous or identical to the polynucleotides summarized in Appendix 1. The homology is preferably greater than 90% and is most preferably greater than 95%.

[00026] Fragments of genes can also be targeted. These fragments are typically in the approximate size range of about 20 nucleotides. Thus, targeted fragments are preferably at least about 15 nucleotides. In certain embodiments, the gene fragment targeted by the RNAi molecule is about 20-25 nucleotides in length. However, other size ranges can also be used. For example, using a *C. elegans* microinjection assay, RNAi "fragments" of about 60 nucleotides with between 95 and 100% identity (to a nematode gene) were determined to cause excellent inhibition.

[00027] Thus, RNAi molecules of the subject invention are not limited to those that are targeted to the full-length polynucleotide or gene. The nematode gene product can be inhibited with a RNAi molecule that is targeted to a portion or fragment of the exemplified polynucleotides; high homology (90-95%) or identity is also preferred, but not necessarily essential, for such applications.

[00028] The polynucleotide sequences identified in Appendix A and shown in the Sequence ID listing are from genes encoding nematode proteins having the functions

shown in Appendix 1. The genes exemplified herein are representative of particular classes of proteins which are preferred targets for disruption according to the subject invention. These classes of proteins include, for example, proteins involved in ribosome assembly; neurol transmitter receptors and ligands; electron transport proteins; metabolic pathway proteins; and protein and polynucleotide production, folding, and processing proteins.

[00029] Genetic regulatory sequences, such as promoters, enhancers, and terminators, can be used in genetic constructs to practice the subject invention. Such constructs themselves can also be used for nematode control. Various constructs can be used to achieve expression in specific plant tissues (by using root specific promoters, for example) and/or to target specific nematode tissues (by using targeting elements or adjacent targeting sequences, for example).

[00030] In a specific embodiment of the subject invention, plant cells, preferably root cells, are genetically modified to produce at least one RNAi that is designed to be taken up by nematodes during feeding to block expression (or the function of) of a target gene. As is known in the art, RNAi can target and reduce (and, in some cases, prevent) the translation of a specific gene product. RNAi can be used to reduce or prevent message translation in any tissue of the nematode because of its ability to cross tissue and cellular boundaries. Thus, RNAi that is contacted with a nematode by soaking, injection, or consumption of a food source will cross tissue and cellular boundaries. RNAi can also be used as an epigenetic factor to prevent the proliferation of subsequent generations of nematodes.

[00031] Nematode polynucleotide sequences disclosed herein demonstrate conserved nucleotide motifs among different nematode genera. Conserved nucleotide motifs strongly suggest that these sequences are associated with viability and/or parasitism and are functionally conserved and expressed in both *Meloidogyne incognita* (root-knot nematode) and *Globodera rostochiensis* and *Globdera pallids* (potato cyst nematodes). The use of these polynucleotides, and RNAi inhibitors thereof, is advantageous because such RNAi can be designed to have broad RNAi specificity and are thus useful for controlling a large number of plant parasitic nematodes *in planta*. Because the genes identified in this disclosure are associated with nematode survival

and/or parasitism, RNAi inhibition of these genes (arising from contacting nematodes with compositions comprising RNAi molecules) prevents and/or reduces parasitic nematode growth, development, and/or parasitism.

[00032] Methods of the subject invention include the transformation of plant cells with genes or polynucleotides of the present invention, which can be used to produce nematode inhibitors or RNAi in the plants. In one embodiment, the transformed plant or plant tissue can express RNAi molecules encoded by the gene or polynucleotide sequence introduced into the plant. Other nematode inhibitors contemplated by the invention include antisense molecules specific to the polynucleotide sequences disclosed herein. The transformation of plants with genetic constructs disclosed herein can be accomplished using techniques well known to those skilled in the art and can involve modification of the gene(s) to optimize expression in the plant to be made resistant to nematode infection and infestation. Furthermore, it is known in the art that many tissues of the transgenic plants (such as the roots) can be targeted for transformation.

[00033] RNA-mediated interference (RNAi) of gene expression. Several aspects of root-knot nematode biology make classical genetic studies difficult with this organism. Since root-knot nematodes reproduce by obligatory mitotic parthenogenesis, the opportunity to perform genetic crosses is not available. Microinjection of RNAi can be used to manipulate gene expression in *C. elegans* (Fire, A., S. Xu, M. K. Montgomery, S. A. Kostas, S. E. Driver, and C. C. Mello. [1998] "Potent and specific genetic interference by double-stranded RNA in *Caenorhabditis elegans*" *Nature* 391:806-811). Microinjecting (into adult nematodes) RNAi can turn off specific genes in progeny worms complementary to the coding region of the genes. Moreover, gene inhibition occurs in progeny when RNAi is injected into the body cavity of the adult, indicating the ability of the RNAi to cross cellular boundaries. This RNAi injection method provides a molecular genetic tool that allows for analysis of gene function in root-knot nematodes.

[00034] RNAi can be taken up by *C. elegans* by simply soaking the nematodes in a solution RNAi. This results in targeted inhibition of gene expression in the nematode (Maeda, I., Y. Kohara, M. Yamamoto and A. Sugimoto [1999] "RNAi screening with a non-redundant cDNA set" International Worm Meeting, Madison, WI, abstract 565). Nematodes fed *E. coli* expressing RNAi also demonstrate targeted and

heritable inhibition of gene expression (Sarkissian, M., H. Tabara and C. C. Mello [1999] "A mut-6 screen for RNAi deficient mutants" International Worm Meeting, Madison, WI, abstract 741; Timmons, L. and A. Fire [1998] "Specific interference by ingested dsRNA" *Nature* 395:854; WO 99/32619, hereby incorporated by reference in its entirety).

[00035] Accordingly, one aspect of the instant invention is directed to the control of nematodes comprising contacting nematodes with compositions comprising RNAi molecules specific to the nematode genes disclosed herein. The contacting step may include soaking the nematodes in a solution containing RNAi molecules, feeding nematodes RNAi molecules contained in microbes or plant cells upon which the nematode feeds, or injecting nematodes with RNAi. Nematodes can also be "contacted" and controlled by RNAi expressed in plant tissues that would be consumed, ingested, or frequented by nematodes.

[00036] The RNAi molecules provided to the nematodes may be specific to a single gene. A "cocktail" of RNAi molecules specific to various segments of a single gene can also be used. In addition, a "multigene cocktail" of RNAi molecules specific to two or more genes (or segments thereof) may be applied to the nematodes according to the subject invention.

[00037] In addition to RNAi uptake mediated by transgenic plants, nematodes can be directly transformed with RNAi constructs of cDNAs encoding secretory or other essential proteins to reduce expression of the corresponding gene. The transgenic animals can be assayed for inhibition of gene product using immunoassays or for reduced virulence on a host. Progeny of affected worms can also be assayed by similar methods.

[00038] Procedures that can be used for the preparation and injection of RNAi include those detailed by Fire *et al.*, (1998; <ftp://ciw1.ciwemb.edu>). Root-knot nematodes can be routinely monoxenically cultured on *Arabidopsis thaliana* roots growing on Gamborg's B-5/Gelrite® media. This nematode-host pathosystem is ideally suited for these microinjection experiments since limited root galling results in the parasitic stages (late J2 through adult females) developing outside of the root for easy accessibility for injecting. Another advantage is the parthenogenic reproduction of root-knot nematodes, which makes fertilization by males unnecessary for egg production. The RNAi can be injected into the body cavity of parasitic stages of root-knot nematodes

feeding on *A. thaliana* roots using microinjection. Control nematodes can be injected in parallel with only buffer or an unrelated RNAi. Injected nematodes can be monitored for egg production, and the eggs can be collected for the assays described below. Female root-knot nematodes will typically survive and lay more than 250 eggs following 1 μ l injection of buffer.

[00039] Alternatively, methods are available for microinjecting materials directly into the plant root cells upon which nematodes feed: giant cells or syncytial cells (Böckenhoff, A. and F.M.W. Grundler [1994] "Studies on the nutrient uptake by the beet cyst nematode *Heterodera schachtii* by *in situ* microinjection of fluorescent probes into the feeding structures in *Arabidopsis thaliana*" *Parasitology* 109:249-254). This provides an excellent test system to screen RNAi molecules for efficacy by directly inhibiting growth and development of the nematode feeding upon the microinjected plant cell, or by reducing fecundity and the ability of said nematode to generate pathogenic or viable progeny.

[00040] There are a number of strategies that can be followed to assay for RNAi gene interference. Inhibition of gene expression by RNAi inhibits the accumulation of the corresponding secretory protein in the esophageal gland cells of transgenic J2 hatched from the eggs produced by the injected nematodes. In the first assay, polyclonal antibodies to the target gene product can be used in immunolocalization studies (Hussey, R. S. [1989] "Monoclonal antibodies to secretory granules in esophageal glands of *Meloidogyne* species" *J. Nematol.* 21:392-398; Borgonie, G, E. van Driessche, C. D. Link, D. de Waele, and A. Coomans [1994] "Tissue treatment for whole mount internal lectin staining in the nematodes *Caenorhabditis elegans*, *Panagrolaimus superbus* and *Acrobeloides maximus*" *Histochemistry* 101:379-384) to monitor the synthesis of the target protein in the gland cells of progeny of the injected nematodes, or in any other nematode tissue that fails to express the essential targeted gene. Interference of endogenous gene activity by the RNAi eliminates binding of the antibodies to secretory granules in the glands, or any other target tissue, of the transgenic nematodes, and can be monitored by these *in situ* hybridization experiments. Control nematodes injected only with the injection buffer can be processed similar to the RNAi treated nematodes.

[00041] Another assay is designed to determine the effect of the RNAi on reducing the virulence of J2 progeny of the injected females. Egg masses from injected females can be transferred singly to *A. thaliana* plates to assess the ability of the transgenic J2 to infect roots. The J2 hatching from the eggs transferred to the plates can be monitored; after 25 days the number of galls with egg laying females can be recorded. The *A. thaliana* roots can also be stained with acid fuschin to enumerate the number of nematodes in the roots. Egg masses from nematodes injected only with the injection buffer can be handled similarly and used as controls. The treatments can be replicated, and the root infection data can be analyzed statistically. These experiments can be used to assess the importance of the target genes in root-knot nematode's virulence or viability. By staining the J2 progeny of the injected females with the antibodies, it can be determined whether RNAi blocks expression of the targeted gene.

[00042] Additional uses of polynucleotides. The polynucleotide sequences exemplified herein can be used in a variety of ways. These polynucleotides can be used in assays for additional polynucleotides and additional homologous genes, and can be used in tracking the quantitative and temporal expression of parasitism genes in nematodes. These polynucleotides can be cloned into microbes for production and isolation of their gene products. Among the many uses of the isolated gene product is the development of additional inhibitors and modifiers. The protein products of the subject polynucleotides can also be used as diagnostic tools. For example, proteins encoded by the parasitism genes, as identified herein, can be used in large scale screenings for additional peptide inhibitors. The use of peptide phage display screening is one method that can be used in this regard. Thus, the subject invention also provides new biotechnological strategies for managing nematodes under sustainable agricultural conditions.

[00043] Antisense technologies can also be used for phytopathogenic nematode control. Antisense technology can be used to interfere with expression of the disclosed endogenous nematode genes. Antisense technology can also be used to alter the components of plants used as targets by the nematodes. For example, the transformation of a plant with the reverse complement of an endogenous gene encoded by a polynucleotide exemplified herein can result in strand co-suppression and gene silencing

or inhibition of a target involved in the nematode infection process. Thus, the subject invention includes transgenic plants (which are preferably made nematode-resistant in this manner, and other organisms including microbes and phages) comprising RNAi or antisense molecules specific to any of the polynucleotides identified herein.

[00044] Polynucleotide probes. DNA possesses a fundamental property called base complementarity. In nature, DNA ordinarily exists in the form of pairs of anti-parallel strands, the bases on each strand projecting from that strand toward the opposite strand. The base adenine (A) on one strand will always be opposed to the base thymine (T) on the other strand, and the base guanine (G) will be opposed to the base cytosine (C). The bases are held in apposition by their ability to hydrogen bond in this specific way. Though each individual bond is relatively weak, the net effect of many adjacent hydrogen bonded bases, together with base stacking effects, is a stable joining of the two complementary strands. These bonds can be broken by treatments such as high pH or high temperature, and these conditions result in the dissociation, or "denaturation," of the two strands. If the DNA is then placed in conditions which make hydrogen bonding of the bases thermodynamically favorable, the DNA strands will anneal, or "hybridize," and reform the original double-stranded DNA. If carried out under appropriate conditions, this hybridization can be highly specific. That is, only strands with a high degree of base complementarity will be able to form stable double-stranded structures. The relationship of the specificity of hybridization to reaction conditions is well known. Thus, hybridization may be used to test whether two pieces of DNA are complementary in their base sequences. It is this hybridization mechanism which facilitates the use of probes of the subject invention to readily detect and characterize DNA sequences of interest.

[00045] The specifically exemplified polynucleotides of the subject invention can themselves be used as probes. Additional polynucleotide sequences can be added to the ends of (or internally in) the exemplified polynucleotide sequences so that polynucleotides that are longer than the exemplified polynucleotides can also be used as probes. Thus, isolated polynucleotides comprising one or more of the exemplified sequences are within the scope of the subject invention. Polynucleotides that have less nucleotides than the exemplified polynucleotides can also be used and are contemplated within the scope of the present invention. For example, for some purposes, it might be

useful to use a conserved sequence from an exemplified polynucleotide wherein the conserved sequence comprises a portion of an exemplified sequence. Thus, polynucleotides of the subject invention can be used to find additional, homologous (wholly or partially) genes.

[00046] Probes of the subject invention may be composed of DNA, RNA, or PNA (peptide nucleic acid). The probe will normally have at least about 10 bases, more usually at least about 17 bases, and may have about 100 bases or more. Longer probes can readily be utilized, and such probes can be, for example, several kilobases in length. The probe sequence is designed to be at least substantially complementary to a portion of a gene encoding a protein of interest. The probe need not have perfect complementarity to the sequence to which it hybridizes. The probes may be labeled utilizing techniques that are well known to those skilled in this art.

[00047] One approach for the use of the subject invention as probes entails first identifying DNA segments that are homologous with the disclosed nucleotide sequences using, for example, Southern blot analysis of a gene bank. Thus, it is possible, without the aid of biological analysis, to know in advance the probable activity of many new polynucleotides, and of the individual gene products expressed by a given polynucleotide. Such an analysis provides a rapid method for identifying commercially valuable compositions.

[00048] One hybridization procedure useful according to the subject invention typically includes the initial steps of isolating the DNA sample of interest and purifying it chemically. Either lysed nematodes or total fractionated nucleic acid isolated from nematodes can be used. Cells can be treated using known techniques to liberate their DNA (and/or RNA). The DNA sample can be cut into pieces with an appropriate restriction enzyme. The pieces can be separated by size through electrophoresis in a gel, usually agarose or acrylamide. The pieces of interest can be transferred to an immobilizing membrane.

[00049] The particular hybridization technique is not essential to the subject invention. As improvements are made in hybridization techniques, they can be readily applied.

[00050] The probe and sample can then be combined in a hybridization buffer solution and held at an appropriate temperature until annealing occurs. Thereafter, the membrane is washed free of extraneous materials, leaving the sample and bound probe molecules typically detected and quantified by autoradiography and/or liquid scintillation counting. As is well known in the art, if the probe molecule and nucleic acid sample hybridize by forming a strong non-covalent bond between the two molecules, it can be reasonably assumed that the probe and sample are essentially identical or very similar. The probe's detectable label provides a means for determining in a known manner whether hybridization has occurred.

[00051] In the use of the nucleotide segments as probes, the particular probe is labeled with any suitable label known to those skilled in the art, including radioactive and non-radioactive labels. Typical radioactive labels include ^{32}P , ^{35}S , or the like. Non-radioactive labels include, for example, ligands such as biotin or thyroxine, as well as enzymes such as hydrolases or peroxidases, or the various chemiluminescers such as luciferin, or fluorescent compounds like fluorescein and its derivatives. In addition, the probes can be made inherently fluorescent as described in International Application No. WO 93/16094.

[00052] Various degrees of stringency of hybridization can be employed. The more stringent the conditions, the greater the complementarity that is required for duplex formation. Stringency can be controlled by temperature, probe concentration, probe length, ionic strength, time, and the like. Preferably, hybridization is conducted under moderate to high stringency conditions by techniques well known in the art, as described, for example, in Keller, G.H., M.M. Manak (1987) *DNA Probes*, Stockton Press, New York, NY., pp. 169-170.

[00053] As used herein "moderate to high stringency" conditions for hybridization refers to conditions that achieve the same, or about the same, degree of specificity of hybridization as the conditions "as described herein." Examples of moderate to high stringency conditions are provided herein. Specifically, hybridization of immobilized DNA on Southern blots with ^{32}P -labeled gene-specific probes was performed using standard methods (Maniatis *et al.*). In general, hybridization and subsequent washes were carried out under moderate to high stringency conditions that

allowed for detection of target sequences with homology to sequences exemplified herein. For double-stranded DNA gene probes, hybridization was carried out overnight at 20-25° C below the melting temperature (Tm) of the DNA hybrid in 6X SSPE, 5X Denhardt's solution, 0.1% SDS, 0.1 mg/ml denatured DNA. The melting temperature is described by the following formula from Beltz *et al.* (1983):

[00054] $Tm = 81.5^{\circ}\text{C} + 16.6 \cdot \text{Log}[\text{Na}^+] + 0.41(\%G+C) - 0.61(\%\text{formamide}) - 600/\text{length of duplex in base pairs.}$

Washes are typically carried out as follows:

- (1) Twice at room temperature for 15 minutes in 1X SSPE, 0.1% SDS (low stringency wash).
- (2) Once at $Tm - 20^{\circ}\text{C}$ for 15 minutes in 0.2X SSPE, 0.1% SDS (moderate stringency wash).

[00055] For oligonucleotide probes, hybridization was carried out overnight at 10-20° C below the melting temperature (Tm) of the hybrid in 6X SSPE, 5X Denhardt's solution, 0.1% SDS, 0.1 mg/ml denatured DNA. Tm for oligonucleotide probes was determined by the following formula from Suggs *et al.* (1981):

[00056] $Tm (\text{ }^{\circ}\text{C}) = 2(\text{number T/A base pairs}) + 4(\text{number G/C base pairs})$

[00057] Washes were typically carried out as follows:

[00058] (1) Twice at room temperature for 15 minutes 1X SSPE, 0.1% SDS (low stringency wash).

[00059] (2) Once at the hybridization temperature for 15 minutes in 1X SSPE, 0.1% SDS (moderate stringency wash).

[00060] In general, salt and/or temperature can be altered to change stringency. With a labeled DNA fragment of greater than about 70 or so bases in length, the following conditions can be used:

Low:	1 or 2X SSPE, room temperature
Low:	1 or 2X SSPE, 42° C
Moderate:	0.2X or 1X SSPE, 65° C
High:	0.1X SSPE, 65° C.

[00061] Duplex formation and stability depend on substantial complementarity between the two strands of a hybrid, and, as noted above, a certain degree of mismatch

can be tolerated. Therefore, polynucleotide sequences of the subject invention include mutations (both single and multiple), deletions, and insertions in the described sequences, and combinations thereof, wherein said mutations, insertions, and deletions permit formation of stable hybrids with a target polynucleotide of interest. Mutations, insertions, and deletions can be produced in a given polynucleotide sequence using standard methods known in the art. Other methods may become known in the future.

[00062] The mutational, insertional, and deletional variants of the polynucleotide sequences of the invention can be used in the same manner as the exemplified polynucleotide sequences so long as the variants have substantial sequence similarity with the original sequence. As used herein, substantial sequence similarity refers to the extent of nucleotide similarity that is sufficient to enable the variant polynucleotide to function in the same capacity as the original sequence. Preferably, this similarity is greater than 50%; more preferably, this similarity is greater than 75%; and most preferably, this similarity is greater than 90%. The degree of similarity needed for the variant to function in its intended capacity will depend upon the intended use of the sequence. It is well within the skill of a person trained in this art to make mutational, insertional, and deletional mutations that are designed to improve the function of the sequence or otherwise provide a methodological advantage.

[00063] PCR technology. Polymerase Chain Reaction (PCR) is a repetitive, enzymatic, primed synthesis of a nucleic acid sequence. This procedure is well known and commonly used by those skilled in this art (see U.S. Patent Nos. 4,683,195; 4,683,202; and 4,800,159; Saiki *et al.*, 1985). PCR is based on the enzymatic amplification of a DNA fragment of interest that is flanked by two oligonucleotide primers that hybridize to opposite strands of the target sequence. The primers are oriented with the 3' ends pointing towards each other. Repeated cycles of heat denaturation of the template, annealing of the primers to their complementary sequences, and extension of the annealed primers with a DNA polymerase result in the amplification of the segment defined by the 5' ends of the PCR primers. Since the extension product of each primer can serve as a template for the other primer, each cycle essentially doubles the amount of DNA fragment produced in the previous cycle. This results in the exponential accumulation of the specific target fragment, up to several million-fold in a

few hours. By using a thermostable DNA polymerase such as *Taq* polymerase, which is isolated from the thermophilic bacterium *Thermus aquaticus*, the amplification process can be completely automated. Other enzymes that can be used are known to those skilled in the art.

[00064] The polynucleotide sequences of the subject invention (and portions thereof such as conserved regions and portions that serve to distinguish these sequences from previously-known sequences) can be used as, and/or used in the design of, primers for PCR amplification. In performing PCR amplification, a certain degree of mismatch can be tolerated between primer and template. Therefore, mutations, deletions, and insertions (especially additions of nucleotides to the 5' end) of the exemplified polynucleotides can be used in this manner. Mutations, insertions and deletions can be produced in a given primer by methods known to an ordinarily skilled artisan.

[00065] The polynucleotide sequences of the instant invention may be "operably linked" to regulatory sequences such as promoters and enhancers. Nucleic acid is "operably linked" when it is placed into a functional relationship with another nucleic acid sequence. For example, DNA for a presequence or secretory leader is "operably linked" to DNA encoding a polypeptide if it is expressed as a preprotein that participates in the secretion of the polypeptide; a promoter or enhancer is "operably linked" to a coding sequence if it affects the transcription of the sequence; or a ribosome binding site is "operably linked" to a coding sequence if it is positioned so as to facilitate translation. Generally, "operably linked" means that the DNA sequences being linked are contiguous, and, in the case of a secretory leader, contiguous and in reading phase. However, enhancers do not have to be contiguous. Linking is accomplished by ligation at convenient restriction sites. If such sites do not exist, synthetic oligonucleotide adaptors or linkers are used in accordance with conventional practice.

[00066] Polynucleotides and proteins. Polynucleotides of the subject invention can be defined according to several parameters. One characteristic is the biological activity of the protein products as identified herein. The proteins and genes of the subject invention can be further defined by their amino acid and nucleotide sequences. The sequences of the molecules can be defined in terms of homology to certain exemplified sequences as well as in terms of the ability to hybridize with, or be amplified by, certain

exemplified probes and primers. Additional primers and probes can readily be constructed by those skilled in the art such that alternate polynucleotide sequences encoding the same amino acid sequences can be used to identify and/or characterize additional genes. The proteins of the subject invention can also be identified based on their immunoreactivity with certain antibodies.

[00067] The polynucleotides and proteins of the subject invention include portions, fragments, variants, and mutants of the full-length sequences as well as fusions and chimerics, so long as the encoded protein retains the characteristic biological activity of the proteins identified herein. As used herein, the terms "variants" or "variations" of genes refer to nucleotide sequences that encode the same proteins or which encode equivalent proteins having equivalent biological activity. As used herein, the term "equivalent proteins" refers to proteins having the same or essentially the same biological activity as the exemplified proteins.

[00068] It will be apparent to a person skilled in this art that genes within the scope of the subject invention can be identified and obtained through several means. The specific genes exemplified herein may be obtained from root-knot nematodes. Genes, or portions or variants thereof, may also be artificially synthesized by, for example, a gene synthesizer.

[00069] Variations of genes may be readily constructed using standard techniques such as site-directed mutagenesis and other methods of making point mutations and by DNA shuffling, for example. In addition, gene and protein fragments can be made using commercially available exonucleases, endonucleases, and proteases according to standard procedures. For example, enzymes such as *Ba*31 can be used to systematically cut off nucleotides from the ends of genes. In addition, genes that encode fragments may be obtained using a variety of restriction enzymes. Proteases may be used to directly obtain active fragments of these proteins. Of course, molecular techniques for cloning polynucleotides and producing gene constructs of interest are also well known in the art. *In vitro* evaluation techniques, such as MAXYGEN's "Molecular Breeding" can also be applied to practice the subject invention.

[00070] Other molecular techniques can also be applied using the teachings provided herein. For example, antibodies raised against proteins encoded by

polynucleotides disclosed herein can be used to identify and isolate proteins from a mixture of proteins. Specifically, antibodies may be raised to the portions of the proteins that are conserved and most distinct from other proteins. These antibodies can then be used to specifically identify equivalent proteins by immunoprecipitation, enzyme linked immunosorbent assay (ELISA), or Western blotting. Antibodies to proteins encoded by polynucleotides disclosed herein, or to equivalent proteins, can readily be prepared using standard procedures known in the art. The genes that encode these proteins can be obtained from various organisms.

[00071] Because of the redundancy of the genetic code, a variety of different DNA sequences can encode the amino acid sequences encoded by the polynucleotide sequences disclosed herein. It is well within the skill of a person trained in the art to create these alternative DNA sequences encoding proteins having the same, or essentially the same, amino acid sequence. These variant DNA sequences are within the scope of the subject invention. As used herein, reference to "essentially the same" sequence refers to sequences that have amino acid substitutions, deletions, additions, or insertions that do not materially affect biological activity. Fragments retaining the characteristic biological activity are also included in this definition.

[00072] A further method for identifying genes and polynucleotides (and the proteins encoded thereby) of the subject invention is through the use of oligonucleotide probes. Probes provide a rapid method for identifying genes of the subject invention. The nucleotide segments that are used as probes according to the invention can be synthesized using a DNA synthesizer and standard procedures.

[00073] The subject invention comprises variant or equivalent proteins (and nucleotide sequences coding for equivalent proteins or for inhibitors of the genes encoding such proteins) having the same or similar biological activity of inhibitors or proteins encoded by the exemplified polynucleotides. Equivalent proteins will have amino acid similarity with an exemplified protein (or peptide). The amino acid and/or nucleotide identity will typically be greater than 60%. Preferably, the identity will be greater than 75%. More preferably, the identity will be greater than 80%, and even more preferably greater than 90%. Most preferably, the identity will be greater than 95%. RNAi molecules will also have corresponding identities in these preferred ranges. These

identities are as determined using standard alignment techniques for determining amino acid and/or nucleotide identity. The identity/similarity will be highest in critical regions of the protein or gene including those regions that account for biological activity or that are involved in the determination of three-dimensional configuration that is ultimately responsible for the biological activity. In this regard, certain amino acid substitutions are acceptable and can be expected if these substitutions are in regions which are not critical to activity or are conservative amino acid substitutions which do not affect the three-dimensional configuration of the molecule. For example, amino acids may be placed in the following classes: non-polar, uncharged polar, basic, and acidic. Conservative substitutions whereby an amino acid of one class is replaced with another amino acid of the same type fall within the scope of the subject invention so long as the substitution does not materially alter the biological activity of the compound. Below is a list of examples of amino acids belonging to various classes

Class of Amino Acid	Examples of Amino Acids
Nonpolar	Ala, Val, Leu, Ile, Pro, Met, Phe, Trp
Uncharged Polar	Gly, Ser, Thr, Cys, Tyr, Asn, Gln
Acidic	Asp, Glu
Basic	Lys, Arg, His

[00074] In some instances, non-conservative substitutions can also be made. The critical factor is that these substitutions must not detract from the ability to manage nematode-caused diseases.

[00075] An "isolated" or "substantially pure" nucleic acid molecule or polynucleotide is a polynucleotide that is substantially separated from other polynucleotide sequences which naturally accompany a nucleic acid molecule. The term embraces a polynucleotide sequence which was removed from its naturally occurring environment by the hand of man. This includes recombinant or cloned DNA isolates,

chemically synthesized analogues and analogues biologically synthesized by heterologous systems. An "isolated" or "purified" protein, likewise, is a protein removed from its naturally occurring environment.

[00076] Recombinant hosts. The genes, antisense, and RNAi polynucleotides within the scope of the present invention can be introduced into a wide variety of microbial or plant hosts. Plant cells can be transformed (made recombinant) in this manner. Microbes, for example, can also be used in the application of RNAi molecules of the subject invention in view of the fact that microbes are a food source for nematodes

[00077] There are many methods for introducing a heterologous gene or polynucleotide into a host cell or cells under conditions that allow for stable maintenance and expression of the gene or polynucleotide. These methods are well known to those skilled in the art. Synthetic genes, such as, for example, those genes modified to enhance expression in a heterologous host (such as by preferred codon usage or by the use of adjoining, downstream, or upstream enhancers) that are functionally equivalent to the genes (and which encode equivalent proteins) can also be used to transform hosts. Methods for the production of synthetic genes are known in the art.

[00078] Where the gene or polynucleotide of interest is introduced via a suitable vector into a microbial host, and said host is applied to the environment in a living state, certain host microbes are preferred. Certain microorganism hosts are known to occupy the phytosphere, phylloplane, phyllosphere, rhizosphere, and/or rhizoplane of one or more crops of interest. These microorganisms can be selected so as to be capable of successfully competing in the particular environment (crop and other habitats) with the wild-type microorganisms, provide for stable maintenance and expression of the gene expressing a polypeptide of interest, and, desirably, provide for improved protection of the protein/peptide from environmental degradation and inactivation.

[00079] A large number of microorganisms is known to inhabit the phylloplane (the surface of the plant leaves) and/or the rhizosphere (the soil surrounding plant roots) of a wide variety of important crops. These microorganisms include bacteria, algae, and fungi. Of particular interest are microorganisms, such as bacteria, e.g., genera *Pseudomonas*, *Erwinia*, *Serratia*, *Klebsiella*, *Xanthomonas*, *Streptomyces*, *Rhizobium*, *Rhodopseudomonas*, *Methylophilius*, *Agrobacterium*, *Acetobacter*, *Lactobacillus*,

Arthrobacter, Azotobacter, Leuconostoc, and Alcaligenes; fungi, particularly yeast, e.g., genera Saccharomyces, Cryptococcus, Kluyveromyces, Sporobolomyces, Rhodotorula, and Aureobasidium. Of particular interest are the pigmented microorganisms.

[00080] Methods of the subject invention also include the transformation of plants or plant tissue with genes which encode the RNAi molecules of the present invention. In one embodiment, the transformed plant or plant tissue expresses antisense RNA and/or RNAi. Transformation of cells can be made by those skilled in the art using standard techniques. Materials necessary for these transformations are disclosed herein or are otherwise readily available to the skilled artisan.

[00081] Additional methods and formulations for control of pests. Control of nematode pests using the RNAi molecules of the instant invention can be accomplished by a variety of additional methods that would be apparent to those skilled in the art having the benefit of the subject disclosure. A "cocktail" of two or more RNAi molecules can be used to disrupt one or more of the genes identified herein. The "cocktail" of RNAi molecules may be specific to segments of a single gene or the entire gene. A "multigene cocktail" of RNAi molecules specific to two or more genes (or segments thereof) is also encompassed by the instant invention. In another embodiment of the instant invention, the disclosed RNAi molecules, cocktails, and/or multigene cocktails thereof, may be used in conjunction with other known nematode control agents and methodologies. Such cocktails can be used to combat the development of resistance by nematodes to a certain inhibitor or inhibitors.

[00082] Compositions of the subject invention which comprise RNAi molecules and carriers can be applied, themselves, directly or indirectly, to locations frequented by, or expected to be frequented by, nematodes. Microbial hosts which were transformed with polynucleotides that encode RNAi molecules, express said RNAi molecules, and which colonize roots (e.g., *Pseudomonas, Bacillus*, and other genera) can be applied to the sites of the pest, where they will proliferate and be ingested. The result is control of the pest. Thus, methods of the subject invention include, for example, the application of recombinant microbes to the pests (or their locations). The recombinant microbes may also be transformed with more than one RNAi molecule thereby delivering a "cocktail" of RNAi molecules to the nematode pests. A carrier may be any substance suitable for

delivering the RNAi molecules to the nematode. Acceptable carriers are well known in the art and also are commercially available. For example, such acceptable carriers are described in E.W. Martin's *Remington's Pharmaceutical Science*, Mack Publishing Company, Easton, PA..

[00083] All patents, patent applications, provisional applications, and publications referred to or cited herein are incorporated by reference in their entirety to the extent they are not inconsistent with the explicit teachings of this specification.

[00084] Following are examples that illustrate procedures for practicing the invention. These examples should not be construed as limiting. All percentages are by weight and all solvent mixture proportions are by volume unless otherwise noted.

Example 1—Production of Hairy Roots for RNAi Testing

[00085] A hairy root assay system was developed for testing the anti-nematode activity of RNAi molecules.

[00086] *Agrobacterium rhizogenes*: Several *Agrobacterium rhizogenes* strains produce hairy roots on a variety of plant species. *A. rhizogenes* strains, A4, 15834, 8196 and LBA4404 demonstrate hairy root development on tomato and sugar beet, with A4 being the most efficient. The *A. rhizogenes* strain K599 demonstrated very efficient formation on transgenic soybean hairy roots and was also effective on sugar beet and *Arabidopsis*. However, stain K599 failed to produce hairy roots on tomato tissues possibly due to hyper-virulence.

[00087] Hairy root production: Transgenic hairy roots were identified by stable GUS expression in tomato, sugar beet, soybean and *Arabidopsis*. The construct pAKK1401 (pNOS / NPT-II / tNOS // pSU / GUS / tNOS) was used to produce hairy roots when transformed into *A. rhizogenes* strains A4 or K599. Transgenic roots were identified by GUS expression.

Example 2 – Protocol for Electro-competent *Agrobacterium* and Electroporation

[00088] Electro-competent Agrobacterium Protocol:

- [00089] 1. Grow *Agrobacterium* overnight in 5 mls LB + antibiotics at 30°C on shaker (for *Agrobacterium rhizogenes* strain K599 no antibiotics are needed).
- [00090] 2. Use the 5 mls of overnight culture to inoculate 500 mls LB + antibiotics at 30°C on shaker. Grow overnight.
- [00091] 3. Add liquid culture in eight 50 ml polypropylene orange cap tubes.
- [00092] 4. Centrifuge 10 min., 4000 rpm, 4°C.
- [00093] 5. Resuspend cells in each tube with 20 mls 10% glycerol (on ice)
- [00094] 6. Centrifuge 10 min., 4000 rpm, 4°C.
- [00095] 7. Resuspend cells in each tube with 10 mls 10% glycerol (on ice).
- [00096] 8. Centrifuge 10 min., 4000 rpm, 4°C.
- [00097] 9. Resuspend cells in each tube with 2 mls 10% glycerol (on ice).
- [00098] 10. Aliquot 50 µl into cold Eppendorf tube and place onto dry ice.
- [00099] 11. Store electro-competent cells at -80°C. These cells can be used for up to two years.

[000100] Electroporations:

- [000101] 1. Add 1 µl to 5 µl of DNA (resuspended in H₂O and not TE or other buffer) to 50 µl of *Agrobacterium* electrocompetent cells and mix.
- [000102] 2. Transfer 20 µl of DNA/*Agrobacterium* mix to cuvette.
- [000103] 3. Electroporate:
25µF, 400 Ω resistance, 2.5 volts (0.2cm cuvette) or 1.8 volts (0.1cm cuvette for BioRad electroporator. 330 µF, 4000 kΩ, low w, fast charge rate for BRL Electroporator.
- [000104] 4. Add 1ml of LB and transfer to Eppendorf tube.
- [000105] 5. Shake at 30°C for 2 hours.
- [000106] 6. Centrifuge down cells (2 min. 14 krpm).
- [000107] 7. Plate all onto LB + antibiotics (most *Agrobacterium* strains are naturally streptomycin resistant).

Example 3 – Protocol for Production of Transgenic Hairy Roots on Soybean

[000108] Seed Sterilization. Rinse the soybean seed with 70% ETOH for 2-5 min. Remove and add 20% Clorox and shake for 20-25 min. Rinse 3X with sterile water. Plate the seed, 5 seed per plate, onto $\frac{1}{2}$ MSB5 + 2% sucrose + 0.2% gel (referred to as $\frac{1}{2}$ MSB5). Place seed into chamber at 25C, 16/8 photoperiod for 5-7 day (depending on genotype) germination period. After 1 week seedlings can be placed into cold room for longer storage if necessary (not to exceed 2 weeks).

[000109] Agrobacterium Preparation. For Agrobacterium rhizogenes strain K599, take a small sample from frozen glycerol into 25-50 ml of NZYM media with 50 mg/L kanamycin in a 125-250 ml Erlenmyer flask. Place onto shaker at 28-30 °C for 16 - 20 hours. Pour sample into centrifuge tube and centrifuge the bacterium at 4000 rpm for 10 min. Pour off supernatant and re-suspend the pellet with an equal volume of liquid $\frac{1}{2}$ MSB5 + 200 μ M acetosyringone. Use pipette to re-suspend the pellet and homogenize the sample (remove all clumps). To determine O.D., prepare a 1:10 dilution by putting 900 μ l $\frac{1}{2}$ MSB5 into cuvette and add 100 μ l of bacterial sample. Determine the O.D.₆₆₀ and calculate the volume needed to adjust (dilute) OD to approximately 0.2 for inoculation. Check final O.D.

[000110] Explant Preparation and inoculation. Place a sterile filter paper onto plates of 1/2 MSB5. Cut soybean cotyledons just above the shoot apex and place onto plate. Lightly scar the cotyledon's abaxial surface (flat side, upper surface that reaches toward sun) with a scalpel blade. Cut each cotyledon transversely into 2-3 pieces (no smaller than 1 cm). Add approximately 10 ml of prepared bacterial solution to each plate and allow cotyledons to incubate for 1 hr. Remove the bacteria using a vacuum aspirator fitted with sterile pipette tip, ensure that there is no standing liquid. Orient all explants with abaxial surface up and wrap plates for a 3 day co-culture, 25°C in light (16/8 photoperiod).

[000111] Hairy root selection and maintenance. After 3 day co-culture, wash explants with liquid $\frac{1}{2}$ MSB5 + 500 mg/L carbenicillin. Transfer the explants abaxial side up to selection media, $\frac{1}{2}$ MSB5 supplemented with 500 mg/L carbenicillin and 200 mg/L kanamycin. Roots should develop in approximately 2-3 weeks. The roots will form primarily from the cut vascular bundles with other roots developing from the small cuts on cotyledon surface. Remove roots (>1cm in length) and place onto replica media with

transfers to fresh media every 2 weeks to prevent *Agrobacterium* overgrowth. After 6-8 weeks on selection the roots can be moved to media without kanamycin, however carbenicillin must remain in media for several months for continued suppression of *Agrobacterium*. At this stage roots can be used for testing RNAi for nematode control. Sterilized nematodes can be added and observed for RNAi affects.

Example 4 – Testing of RNAi for Plant Parasitic Nematode Control.

[000112] Various types of nematodes can be used in appropriate bioassays. For example, *Caenorhabditis elegans*, a bacterial feeding nematode, and plant parasitic nematodes can be used for bioassay purposes. Examples of plant parasitic nematodes include a migratory endo-parasite, *Pratylenchus scribneri* (lesion), and two sedentary endo-parasites, *Meloidogyne javanica* (root-knot) and *Heterodera schachtii* (cyst).

[000113] *C. elegans*: RNAi vectors can be tested through expression of the RNAi in *E. coli*. *C. elegans* are fed *E. coli* and assayed for their growth by measuring growth of nematodes, production of eggs and viability of offspring. Another approach is to inject dsRNA directly into living nematodes. Finally, soaking nematodes in a solution of *in vitro*-prepared RNAi can quickly establish efficacy of treatment.

[000114] *P. scribneri*: The *P. scribneri* *in vitro* feeding assay uses a corn root exudate (CRE) as a feeding stimulus and both the red dye Amaranth or potassium arsenate as feeding indicators. Feeding is confirmed after seven days by the presence of red stained intestinal cells in live worms exposed to the Amaranth or death of worms exposed to arsenate. This bioassay is used to test soluble toxins or RNAi. *P. scribneri* has also been cultured on wild type roots of corn, rice and *Arabidopsis*, and on *A. rhizogenes*-induced hairy roots of sugar beet and tomato. *P. scribneri* is very valuable in evaluating transgenic hairy roots because of the non-specific feeding of these worms.

[000115] *M. javanica*: Nematode eggs are sterilized using bleach and are used to inoculate hairy roots expressing RNAi. Nematodes are assessed for their growth by measuring knots, egg masses or production of viable eggs. An alternative approach is to microinject dsRNA directly into root feeding sites or into living female nematodes.

[000116] *H. schachtii*: Cultures of this nematode were maintained on sugar beets. Nematode eggs are sterilized using bleach and used to inoculate hairy roots

expressing RNAi. Nematodes can be assessed for their growth by measuring knots, egg masses or production of viable eggs.

Example 5 – Plant Expression Vectors for RNAi

[000117] Modular Binary Construct System (MBCS): An important aspect of the subject disclosure is the Modular Binary Construct System. The MBCS eases the burden of construct development by creating modular pieces of DNA that can be easily added, removed, or replaced with the use of low frequency cutting restriction enzymes (8-base cutters). These constructs are useful for delivery of a variety of genes to plant cells and is not limited to the delivery of RNAi genes. To develop this system; a series of six, 8-base cutter restriction enzyme sites was placed between the left and right Ti borders of a previously created kan^R/tet^R binary plasmid (Figure 1). The production of both kan^R and tet^R MCBS aids the testing of constructs using different strains of *Agrobacterium rhizogenes* in different plant species. In addition to the MBCS, a series of shuttle vectors were created that aid in the cloning of useful DNA fragments by containing the multi-cloning site (MCS) of a modified Bluescript plasmid flanked by 8-base restriction sites (Figure 2). With six 8-base cutter sites, each site is, preferably, reserved for a particular function (Figures 3 and 4). Because of the close proximity of the *Pme* I and *Sgf*I sites to the left and right border of the binary vector, these sites are, preferably, reserved for gene tagging and enhancer trap experiments. The *Not* I site is, preferably, reserved for plant selectable markers (Figure 5). The *Pac* I site is reserved, preferably, for Plant Scorable Markers (Figure 6). The *Asc* I site is, preferably, reserved for RNAi experiments (Figures 7 and 8), while the *Sbf*I site is, preferably, reserved for anti-nematode proteins. The restriction sites that are denoted in the Figures are, preferably, reserved for the denoted insertions; however, the MCBS binary and shuttle vectors do not require the restriction sites to contain these suggested inserts.

[000118] Plant Selectable Markers for MBCS: To further develop the MBCS, a series of plant selectable markers were added to the MBCS (Figure 5). Plant selectable markers that were added to the MBCS include: pNOS/NPT-II/tNOS (kan^R), pNOS/Bar/tNOS (basta^R for dicots), pUBI/Intron-Bar/tNOS (basta^R for monocots), and pUBI/Intron-PMI/tNOS (mannitol isomerase^R).

[000119] Reporter Genes for MBCS: Four exemplary reporter genes are used in the MBCS are provided in Figure 6 and Appendix 2. GUS, a nuclear localized GUS, GEP, and the anthocyanin transcriptional activator *papIC* genes into the MBCS.

[000120] Promoters for MBCS: We cloned several useful constitutive and nematode-inducible promoters (Figures 6, 7 and Appendix 2). Constitutive promoters include the SuperUbiquitin promoter from pine (pSU) and two promoter regions from the Strawberry Banding Vein virus (pSBV₁ and pSBV₂). Seven nematode-inducible promoters from *Arabidopsis* were also been cloned.

[000121] The following Scorable marker clones have been constructed and placed in the MBCS, NPT-II binary vector (pNOS/NPT-II/tNOS):

Intron/GUS/tNOS	Intron/NLS-GUS/tNOS	Intron/GFP/tNOS
pSU/Intron/GUS/tNOS	pSU/Intron/NLS-GUS/tNOS	pSU/Intron/GFP/tNOS
pSBV ₁ /Intron/GUS/tNOS	pSBV ₁ /Intron/NLS-GUS/tNOS	pSBV ₁ /Intron/GFP/tNOS
pSBV ₂ /Intron/GUS/tNOS	pSBV ₂ /Intron/NLS-GUS/tNOS	pSBV ₂ /Intron/GFP/tNOS
pKT/Intron/GFP/tNOS		
pKA/Intron/GFP/tNOS		

Example 6 – Control of Plant parasitic nematodes using RNAi in planta

[000122] Production of RNAi Vector. The RNAi shuttle vector to be used is adapted from the Modular Binary Construct System (MBCS - See Example 5). RNAi shuttle vectors preferably comprise a promoter, intron, antisense RNAi, stuffer fragment, sense RNAi, and terminator (See Figures 7 and 8 and Appendix 2 for more details). The plant promoter can be constitutive, tissue-specific or nematode-inducible. The intron is necessary to eliminate expression in *Agrobacterium*.

[000123] The anti-sense and sense RNAi molecules comprise nematode-specific sequences and are disclosed herein. These genes are associated with pathogenesis, growth, or other cellular function in nematodes. An exemplary group of RNAi sequences for use in plant/nematode control may be based upon:

[000124] 1. Genes specific for nematode esophageal gland cells.

[000125] 2. Genes specific for plant parasitic nematodes but not other free living nematodes.

- [000126] 3. Genes common to all plant parasitic nematodes.
- [000127] 4. Genes common to all nematodes (nematode-specific).
- [000128] 5. Genes specific for important tissues or cell types.
- [000129] 6. Genes from large gene families.
- [000130] 7. Genes involved in nematode signal transduction or other cellular pathways.

[000131] Appropriate RNAi constructs allow for the formation of dsRNA molecules (the sense and antisense strands join to form the dsRNA). The terminator sequence adds a poly-A tail for transcriptional termination. The RNAi shuttle vector can then be subcloned into the MBCS and transformed into *Agrobacterium rhizogenes*.

[000132] Plant Transformation with RNAi Vectors. An exemplary transformation system for generating hairy roots using *Agrobacterium rhizogenes* is provided below. The RNAi vector once introduced into the MBCS can subsequently (as a binary vector) be transformed in *A. rhizogenes* using, for example, the electroporation protocol of Example 2. Once the *A. rhizogenes* is confirmed to contain the plasmid, it is then used in generating hairy roots (See Example 3). Using this protocol transgenic hairy roots expressing RNAi are isolated, cultured and tested.

[000133] Testing of RNAi Vector for Nematode or Plant Pathogen Resistance. RNAi expressing hairy roots can be inoculated with sterilized nematodes. Infested hairy roots can be observed and the effect on nematodes determined. An alternative approach involves the microinjection of RNAi directly into root feeding sites (giant-cells for root-knot nematode, and syncytia for cyst nematodes) or into living female nematodes.

Example 7 – Insertion of Genes Into Plants

[000134] One aspect of the subject invention is the transformation of plants with genes encoding proteins of the present invention. Transformation of plants as described herein can be used to improve the resistance of these plants to attack by the target pest.

[000135] Genes, polynucleotides, and/or RNAi molecules as disclosed or suggested herein can be inserted into plant cells using a variety of techniques which are

well known in the art. For example, a large number of cloning vectors, for example, pBR322, pUC series, M13mp series, pACYC184, pMON, etc., are available for preparation for the insertion of foreign genes into higher plants via injection, biolistics (microparticle bombardment), *Agrobacterium tumefaciens*, or *Agrobacterium rhizogenes*-mediated transformation, or electroporation as well as other possible methods. Once the inserted DNA has been integrated into the genome, the genetically modified-cell(s) can be screened via a vector carried-selectable marker that confers on the transformed plant cells resistance to a biocide or an antibiotic, such as kanamycin, G418, bleomycin, hygromycin, chloramphenicol, or bialaphos, *inter alia*. The transformed cell will be regenerated into a morphologically normal plant. The transgene(s) in the transgenic plant is relatively stable and can be inherited by progeny plants.

[000136] If a transformation event involves a germ line cell, then the inserted DNA and corresponding phenotypic trait(s) will be transmitted to progeny plants. Such plants can be grown in the normal manner and crossed with plants that have the same transformed hereditary factors or other hereditary factors. The resulting hybrid individuals have the corresponding phenotypic properties.

[000137] It should be understood that the examples and embodiments described herein are for illustrative purposes only and that various modifications or changes in light thereof will be suggested to persons skilled in the art and are to be included within the spirit and purview of this application.

We claim:

1. An RNAi molecule, optionally comprising a linker, wherein at least one strand of said RNAi is encoded by a DNA sequence selected from the group consisting of SEQ ID NO: 1 through SEQ ID NO: 139.
2. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 1.
3. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 2.
4. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 3.
5. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 4.
6. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 5.
7. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 6.
8. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 7.
9. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 8.
10. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 9.

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11. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 10.

12. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 11.

13. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 12.

14. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 13.

15. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 14.

16. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 15.

17. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 16.

18. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 17.

19. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 18.

20. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 19.

21. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 20.

33

22. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 21.
23. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 22.
24. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 23.
25. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 24.
26. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 25.
27. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 26.
28. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 27.
29. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 28.
30. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 29.
31. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 30.
32. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 31.

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33. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 32.

34. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 33.

35. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 34.

36. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 35.

37. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 36.

38. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 37.

39. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 38.

40. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 39.

41. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 40.

42. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 41.

43. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 42.

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44. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO:
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45. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO:
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46. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO:
45.

47. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO:
46.

48. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO:
47.

49. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO:
48.

50. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO:
49.

51. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO:
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52. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO:
51.

53. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO:
52.

54. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO:
53.

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55. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO:
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56. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO:
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57. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO:
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58. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO:
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59. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO:
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60. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO:
59.

61. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO:
60.

62. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO:
61.

63. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO:
62.

64. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO:
63.

65. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO:
64.

66. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 65.

67. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 66.

68. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 67.

69. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 68.

70. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 69.

71. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 70.

72. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 71.

73. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 72.

74. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 73.

75. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 74.

76. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 75.

77. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO:
76.

78. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO:
77.

79. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO:
78.

80. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO:
79.

81. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO:
80.

82. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO:
81.

83. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO:
82.

84. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO:
83.

85. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO:
84.

86. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO:
85.

87. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO:
86.

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88. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 87.

89. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 88.

90. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 89.

91. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 90.

92. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 91.

93. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 92.

94. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 93.

95. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 94.

96. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 95.

97. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 96.

98. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 97.

110. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 109.

111. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 110.

112. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 111.

113. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 112.

114. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 113.

115. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 114.

116. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 115.

117. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 116.

118. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 117.

119. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 118.

120. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 119.

121. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 120.

122. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 121.

123. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 122.

124. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 123.

125. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 124.

126. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 125.

127. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 126.

128. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 127.

129. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 128.

130. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 129.

131. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 130.

132. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 131.

133. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 132.

134. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 133.

135. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 134.

136. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 135.

137. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 136.

138. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 137.

139. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 138.

140. An RNAi molecule according to claim 1, wherein said DNA sequence is SEQ ID NO: 139.

141. A transgenic plant or transgenic plant tissue comprising an RNAi molecule according to any of the preceding claims.

142. A method of disrupting cellular processes in a nematode comprising the steps of:

- (a) providing a composition comprising a compound according to any of the preceding claims; and
- (b) contacting a nematode with said composition.

143. An isolated promoter comprising the following nucleotide sequence:

aacagcccaagataacagaaaaagtcaaagggtttcgaaa
gaccacttgtgactaaggatcattcatccataattatctggtagca
cagactcatgataactgcgaggaacacaagttctttacagtgcattc
aaagacacttctcttacggttcattgaaggagccgaccgcagaat
atgtcagagaagctttcactgtgggtaatttattatctatcca
ggtaaaaacctcaaggagatctctttccaaaagacctctacag
ggcaatcaaaaactacagaaccagagttgttagtgcacagagtagac
caatctacctgagaatcacgagttcacccatggtagtggaaaatgat
gacatccttattccataccactggattgaggttaggactatccatgg
aaaaattccatggacaagtcatataagaagaccccaacagtgcagt
atcttccagagataactgcactcagacctaaaaggataaaagcagta
tataatcagtgtactaagatcttcgcagattcaaagaagaagcttaa
ctatgctgatgacaagataattctaataagcaattattcagaattaa
tcaaggagaaaagaattaataactcttcagaatatgaagcccctt
acaagtggccagcttagctactgaaaagacagcaagacaatgg
tctcgatgcaccagaaccacatcttcgcagcagatgtgaagcagcca
gagtggtccacaagacgcactcagaaaaggcatcttctaccgacaca
gaaaaagacaaccacagctcatccaaacatgttagactgtcgat
gcgtcggtcaagataagactgaccccaaggccagcactaaagaagaa
ataatgcaagtggcttagctccacttttagcttataattatgttt
cattattattctctgtttgtctctatataaaagagcttgcatttt
cattgaaggcagaggcgaacacacacacacagaacacctccctgttaca
aaccatgtatttagctaaacacctttaggag .

144. An isolated promoter comprising the following nucleotide sequence:⁴⁵

tggtgtggacaatggatccggctcgcttagcaacaaggctg
aaaaagattaaacagaaacctgtgatcattagcgttgaccaccacc
aaaacctcctgagccaccaaagcctccagagcctgaaaaacccaaagc
ctccaccagcacctgaaccaccaaagcatgtatgcaagccaccc
tgcaacagttgtgatgtgtctgttactacctatgaaagtggaaag
cggctgcaccattctttgagtcataatcgcttaccatagccttcat
gttaagtccctgtattttagccaataactaattcatcatgttctcatgct
ttttgttatttcttcaaataatgaaatctctgttgc
ctcccctgttataatttagtcgttgcatttgcacaagaagtctcatg
agttcatgctaaagaaaataaaagttcaattaaacacccaaatg
tgattaatttccataaacctgtgaagcagaaagtttagtcatttgc
ctgaacagagcttagaagtccattgaaggacatatctcaagtgc
ttgggtcgtagcactcttaggcccattacttcatttgc
attatgcaaaacaagaaatgagacatatggaaacatttgc
cagaaaaaaatagaaaaagcagggacaactaaacaaaaattcagaa
acaagaggcaagtggacgaccacggcgtaagatcaacatgtgg
gtgcatttgcatttgcatttgcatttgcatttgcatttgcatttgc
gttttttttatgtttgttgcatttgcatttgcatttgcatttgc
ctttttataggatagtaaaaaatatgatttatgttgc
cattttgagttaaacctaaacttataacttgcatttgcatttgc
tttcctatacgacatctatcaacatgcatttgcatttgcatttgc
gatgaaactacttgcatttgcatttgcatttgcatttgcatttgc
ttaaatttagtagttgttgcatttgcatttgcatttgcatttgc
aaatcaaaacagtatcgtaacttaggaaatgtttatcg
gtttcaacacatgattgcatttgcatttgcatttgcatttgc
cataacaatcatcactcgtaatatcaaaatgtttatcg
aaagggttatgatttccaaactgcatttgcatttgcatttgc
cacacgtatgcatttgcatttgcatttgcatttgcatttgc
ttgggagagatcataatttgcatttgcatttgcatttgcatttgc
cgtaaaatgaaaatttgcatttgcatttgcatttgcatttgc
ctagttatctttttatataacaattcatgaaatgttgc
tttatacacatcatccaaatgcatttgcatttgcatttgc
caggatagagccaaataagatatacaatccaaatggacc
atgtgctaattcatacatgcatttgcatttgcatttgcatttgc
atgctgagcgttttaagtgtgactaagatctgatgc
aagatggcttttgcatttgcatttgcatttgcatttgc
gattcaattttaaaattctaaataaaaactccaaacc
catgaaacttttttagaaaatccatttgcatttgcatttgc
tgcttgcatttgcatttgcatttgcatttgcatttgc
ctcagaaaaagccatttttttgcatttgcatttgcatttgc
tactgtgcgttctacaaagttgttgcatttgcatttgc
actcacagtcacagagatctgtttgcatttgcatttgc
ttctttccagt.

145. An isolated promoter comprising the following nucleotide sequence:

agcaaagcaagaacaccagagaagaagaaaagcactacaga
gaaaaatgtgagcttaagcgctctccaacaacacttctctgggagtc
taaaggatgctgaaaaagccttggtggtgagacttccgcatattc
caagcatgggttatttttttagcacacaaaactatctgaccctcga
cttggatttcttctgcagttgtccaactacattgaaacggatatg
caggcaacatggatcatgaggtggccatctcgtaagatataacaaag
tgaacaggtcactaaggaaaatacagacggtactggactcggtccaa
gtgttagaaggaggactaaagttcgactcagcaactggcgaattcat
tgcagttagaccttttattcaagaaattgataccaaaagggtctgt
cgtctcttgataatgatgcacatgcaagaagaagtcaggaggatatg
cctgacgatacttcattcaagctccaggaagctaaatctgtcgacaa
tgccattaagtttagaggaggatacaccatgaatcaagaagaccag
gtaagaacttctctatccataaaccatagatggagcgttagaatct
taatccatttcagtttgcaggatcattcatggaggtaatgcta
gtggtcagccatgggcttggatggccaaagagtctggcttgaatggc
agtgaaggaaataaagagcgttgcacttaagctctgtggaaatttc
agatggaatggatccaacaatccgatgcagtggcagtattttgaac
ctaaccaatccatgtcatgcagcatcagattcatcaaattggctca
ggcgcagttctgcgtggaaagctcatctacttccatggaaagattggaa
ccaaatgagaaccacaaacagtaatagcagcggagactggatcaacaa
cgctgatcgtaaaggccagttatagagaagacactgtacgtttcaag
ttcgagccatcagttgggtgtcctcaactctacaaagaagtggaaa
acgttttaaactgcaggacgggctcgttcagctgaagtacttggatg
atgaagaagaatgggtgatgctggttacagattctgatctccaagaa
tggtttggagatattacatggatggaaaacactcggtgaagttct
cgttcgtgattgtctgcccctctaggttagttctgtggcagtaatg
gttatcttggAACAGGCTATGACGTCGTAAGACATAGACACACACA
gttatgtattcccaactgtggatgttttttttttttttttttttttt
gtatgcttataaataggcatgaaggagaagacaattttggatagt
ggagttcagcagaaaatgtatatgttttttttttttttttttttt
agaataaaagtggatgttatatctacgttgctaatgttgcacactgc
tcacccatcttcatataagaaaagagaacacttttagttatccctg
tgatgcagaatcgatttttttttttttttttttttttttttttttt
aacaagtcactaaatttccgtttatgggtgggttttttttttttt
cgaggactgttttttttttttttttttttttttttttttttttttt
ttgggttt
tt
tt
tt
aaaagagggagaagatgcgaagacagaatttcataatttggaaagggt
tcgatatcgatattggaaacgaatcaaggtcaaaaaactcagtcta
atagttgaaatttataatttattatcaatccgattgggtttcgt
tttggttatgggttcgttctatcatcaaaaccaatcggtttgggtcct
aaagataattataatttaccaacaccagtgttaaacaacatataca
acaaacactaaagttagataaacaacaaagaga

146. An isolated promoter comprising the following nucleotide sequence:

48
147. An isolated promoter comprising the following nucleotide sequence:

48

:tggcaaactgagatataagagggaaagggtgatttcatgaa
atttttttttatTTTTTGAATGAATGCAAAATTATTCAAAA
aaaaaaaacTGGCTACATCAAGTACTTCATTCTGAGTTGAA
aatCTAAAGACAACAAAAGACTTACAATTAAATAAAAAATAATAA
aaATACTTATCCTACGAAATTGTTGATTAATAACGTATCT
CTTGGTAAAACAGCGTTATTGACGAAATTGTTATAATGAATAA
aatGATAATAGAAAACAGTGTGGTACGTTAAACCTCTCATTGGC
AAAATAACCGTTATGTATCATGAGTATTGCTACGACAGCGTGCTTA
aatAGTGTGCTTCAGGAGAAAATATAACCAAGTTATTGCTGAAA
ttaccacgcaaATCTGAGGTTGCAATGGCAAAATAAAAAACCAATGT
catttccttaATGTATTAGGTCAATTAAATAAAATTGTACACTTT
ttcacCTGTaaAGCGTTCAAAGTGTAGAATGGATAACTAGAAGGGTC
aaAGGTATAATATTAAATAAGCGAACTCACTTTGCCAAGTGTATT
cacttcttacatttgctgtatAGTTACCCAAAAGTGTATATAT
tcccttatacaATTGTTCTATTCTGGATTATAAGGGGAATAAGAA
aaaAGAAAAGAGAGAGTATATAATAACTTTATAAAGTGTATT
gattctaATTGTAACGAAAAGTCAAGTGAAGAAAACGAAA
agTTTTCTGTTGTTATATCTATAGCCAAGAAAGTTCTCAGA
tttacaAGAAGTAACTGAGAAAACAAAAAAACTTATGAAGCA
tgaaAGACTAATTACGAGGTGATTAATTGAGACAAATTAAACAT
cgaattAAAAGTAACATTGGAGGGTTATATGTTATATATGTGACA
tgataAGTCCGATTCACTGACTAATGTATATCTGGAATCTAACATGG
agaATAGAGAACGAAAGCAGAGCCAAGGTCAACTTGGCCAGACACGAAT
caACAGATTGTGAATGAGACCAATCAATGGTCATAAACCGGTTGGG
tttAAACCGGCAAGTCATCCTGGCTCAATTCCATTGTTATTCTT
catGCAAGACCCCTGTACATAACCAAGACTCCATTACAATATTCT
ttcgtcacgagctacttatttcaATGTGTTACCTCTTCGTGAC
tcttGTTGTTGGTAAAGCCTAGTCAGATGTGTCGGTATATATA
ggcatacatatacaaATGCGACAAAATAAGTATATTATATTGTTAA
tttctatattccatttctatATGATGGCTGGGATTTGACCAAAA
ccctaattcaAGAATAGAATCCAAGATGGGATCAAAGAATATAAT
ctaATGGGCTGACCACATTTCGATTAAATTGCGATAGTTAATT
ctttccactactttatGCCGcAGAAATTGTAATTAGTAAGACAAA
gaaATACAGATATAAGATGGTCGTAGAAACCAGTAGAGGAATTCT
tttctgtggataAGTGGATATTAAAGAGAAATGGTCTTACTCT
TACAGGGAAATGGGAAATAGTAGCCCATTATAATTCTACAGATT
TATATATGATGTTGTATAAGCTAAATAACGTTAAGCATT
ttcaaaaaAAATTACAAGTTCTAGAGACTCTTAAACGTCGGCAATT
TATATTCTACTTTACATGACACTTCAGGAAAAGAAAACATAACTCA
CTAGCAGATCATTAAATTCTTTCTTTTTGAAATGAACCTTAG
TTGTGGTTTATTGTTAGCTAGAAACTTCAGTGTTTTTCC
GCCAATGGTAGTGTGTTGATGGTCCGG .

148. An isolated promoter comprising the following nucleotide sequence:

caatcaaggtaacgaaggaggatcagcgaaaggatgggcta
tatttggagttttcctgcgttaagtaatgcgttatgtatcttcca
tgcggacatataactgaagaataaaactcaactcattgtgttctggtg
tgtttcttctgatcagattcctcggtcatctgcactttctgtgt
gggggctttatataaaaacaagagtagagcgtgtggtaatcttcat
atcttctacaattccactccattctctaattattctcacgtga
tatacacacactcaatcactgatgtactcgatggatgcagcgtgga
actgatgcattgccggggatgtcacttctatcggcttactagaaac
tgtaagtattacaagaaaactcaaaaggattccattatgcaaaaatc
taagagaaagctcactgtggtcttggttacaattatggatctc
aagagacaaatgtatgtaagctaattgatttggtcttgataaaca
ggtagtggaaagtggacaaagctactcaagaactgaagacatcaaca
atgctttgccaatgaagtctcatgggaccgctctccgcacatcttct
actcaagcgacacaacaacacagagaccaagtgaaagaacatatggtgc
gatctaatttgcatagtgcctcacaagaggactgttcaagccat
ggtaggcacgcttgcattctgcgatattctggatttgttgcatttgcatt
tttattttcaccttctagaaagaggtcaaaaagttaatagcttac
cgtgagaatgtgtttcaccagattcatgtgctatgatagaaaaag
acaaaagcaacaagagttttcttgcttaggttacaagaacaaga
gtatcgttataaaagtcaacaaagattgaaacatattttgtcaaggg
agtggtagaatcttccactctttgccttctcaactaagacaa
aaaaaaagacttggactttgtctaagggtttggatattattaacca
agtccctttgcacaaaagtaatattgttttgcattccttttag
aatttagttaatcttaggcttataattggttattacttcttgaaaaa
atgatctgttattcttattcatacttggttacctcgctttatctt
acttctacaaaaggattatcagtgaaagtttagtcttactctcacc
ttccgaaaaataaaacaaaaatatcgatacttcttagatcaaaccaagt
tgattaaaacatccctattccctacgattctgatcttgcatttatt
atcatgttaagatctaaattgacaagaaaactgattttcatcttca
gttagaaaaataattactatttagtgcattgtgcaccgtaaaga
ggtagtttagttactctccatcttgcatttttttttttttttttttt
gaaattatataattaaacatcaatattgttttttttttttttttttt
ggttttatgttttagaaaattccaatatttatatttttttttttttt
agaagcttattcttcaattatttttttttttttttttttttttttt
aaaaatataaaagtctaaatattaaaactcagttgcatttttttttt
cctctccaaagtctccaaagtccaaatatttttttttttttttttt
aaaaggtttatttagtccaaacttagcatgcatttttttttttttt
caagcatt
aatt
tttagcttataaccactccgttttttttttttttttttttttttt
taaatacgttcttcttccctcttatttttttttttttttttttttt
tcttctcatttccatcatcatcatcatcatcatcatcatcatcatcat

149. An isolated promoter comprising the following nucleotide sequence:

atgttgtgagtgaaggagaagaagagggaaacaaagggtatt
tattttagcagttttgtttgtacgcgggtttgtctgtttcaaa
tggtgacgaaacgagtgagagagtgtctgttattaaagaaaaccct
aattaagttagccccgggttataaaaatagtcaaaaagttaggaaa
acgcgtgtgtgagtgagacagagacagccattgttgcattatggg
cttataagcgagacgtgttaattgggtttcctttagggaaa
acaaaagaaaacgtcgccctgagagattcaactctcgccggcagagcc
catgtacttagcaggcacgccttaaccactcgccaaagcgactt
gttgctatgagtttagacaaaatcattaaaattcttattatgatttc
tcatagtgtgtgttatattgtggatctactaaaaattcttgcatt
tattactttatattgtgaatttagttgatataggtaagtacaaaagtt
aactttattattactcaaaattttagatcaactgattttat
gttcccttggtatatacgactatagtttagaaaaaccataa
gattcccttatattcatagatgtgaagagatgagatgagatcttggc
tggagaagaataagttccacgaggaggactttttttggta
agacgaggaggaggactttgggtgatccagtcttacgttagacat
cgaccctacatttattgccttctatcaacatggcaggtaaaa
atcttcattcaaccgaaccaaccaaggcttcccaataatattca
agcaccatccttggaaactcatacataactacagctacactctt
cattttcttcaacgctcaacttaacaaatgatatagtcttagttgc
aattatatgttttaatttagtggttcacatcaaattctgggtttagata
tttgatgactatttcggaaacatctcaatgtccgcaaatacatac
gtctatcatatataatccgtacgttgcattttatagatagaataa
tatggcgtgatcttataatataacatataatagaatcgtagattt
tttattttatattatatacgatataattgcacaaatacttataat
gttggatataatgataccatatttagttacttttttttttttttt
gcgataatataatataacttttataacaaaaatgtataacac
atggtaaagaaaaataaaaatgaagacatgggtgacacgaaaatgg
caactaaatatacatataatagatagactacaatatcccatcataca
caacttttaattgactaatacataacttacacacttttttaattga
ctaattcataacttttatcattgtcaacatgcaaaattcatattcc
gttgaactattattcttatttttttttttttttttttttttttt
aataaaaaatatgatttccaaatgacgttagagcaaaaaaaaaaag
gttgcgtggctggtaaaatgaaaaagcaacgcgttgcatttttt
aagtaatataactgcctctaatttgcatttttttttttttttttt
tctccacttttgcctttcgaaacccctaaaccagaagcaccagat
ttttcaacttttcccgagaacaatagaaaaaccacacttgc
tcttaggttttttttttttttttttttttttttttttttttttt
tcattttggaaagcttacccaccagcgaaaaaaattataacttccatcg
attccctggcttctctctcgctctctgtcatgtgctaaatcgccg
gactgatcctcactgtcacctctgtt .

51

150. An isolated promoter comprising the following nucleotide sequence:

52
151. A transgenic plant or transgenic plant tissue comprising an isolated promoter according to any of claims 143 through 150.

54
APPENDIX 1

SEQ ID NO:	INTERNAL IDENTIFIER	FUNCTION OF POLYNUCLEOTIDE / GENE
1, 2, 3	2293133	glyceraldehyde-3-phosphate-dehydrogenase
4, 5, 6, 7	7143495	Histone H4
8 & 9	7143515	ATP dependent RNA helicase, mRNA sequence
10, 11, 12, 13	7143527	nematode specific
14 & 15	7143602	protein serine-threonine phosphatase 1, catalytic subunit
16 & 17	7143612	40S ribosomal protein S4
18	7143666	cytochrome p450
19, 20, 21, 22	7143675	Neuroendocrine protein 7B2
23, 24, 25	7143839	nematode specific
26	7143863	40S ribosomal protein S17
27 & 28	7144016	vacuolar ATP synthase subunit G
29	7144025	malate dehydrogenase
30 & 31	7144060	J2 pcDNAII Globodera rostochiensis cDNA similar to Bystin, mRNA sequence
32 & 33	7144225	similar to arginine kinase
34	7144354	pyrroline-5-carboxylate reductase

SEQ ID NO:	<u>APPENDIX 1 (cont.)</u> INTERNAL IDENTIFIER	FUNCTION OF POLYNUCLEOTID E / GENE
35, 36, 37, 38	C10	ribosomal protein L18a
39, 40, 41, 42, 43	C118	ribosomal protein S11
44 & 45	C122	ribosomal protein L16/L10E
46 & 47	C127	FMRFamide-related neuropeptide precursor
48	C129	ADP-ribosylation factor 1
49	C130	ribosomal protein L11
50	C137	nematode specific; conserved in <i>C.elegans</i>
51 & 52	C138	ribosomal protein L7
53	C145	ADP/ATP translocase
54 & 55	C148	troponin
56 & 57	C154	calponin
58	C16	translation elongation factor EF1A
59 & 60	C18	40S ribosomal protein S16
61	C27	ubiquitin
62 & 63	C46	nematode specific
64, 65, 66	C48	ribosomal protein S3AE
67	C59	40S ribosomal protein S5/S7

SEQ ID NO:	APPENDIX 1 (cont.) INTERNAL IDENTIFIER	FUNCTION OF POLYNUCLEOTID E / GENE
68	C8	glyceraldehyde 3-phosphate dehydrogenase
69 & 70	C82	60S ribosomal protein L30/L7E
71	C90	glyceraldehyde 3-phosphate dehydrogenase
72	C135	nematode specific
73 & 74	C206	predicted troponin
75	C227	cytochrome P450
76	C238	vacuolar ATP synthase subunit G
77	C246	40S ribosomal protein S4
78	C308	FMRFamide-like neuropeptide precursor
79	C342	ubiquitin
80 & 81	C344	nematode specific; conserved in <i>C.elegans</i>
82, 83, 84, 85	C370	40S ribosomal protein S5/S7
86	C426	nematode specific
87	C458	histone H4
88 & 89	C481	ribosomal protein L30E
90 & 91	C556	nematode specific; conserved in <i>C.elegans</i>

SEQ ID NO:	APPENDIX 1 (cont.) INTERNAL IDENTIFIER	FUNCTION OF POLYNUCLEOTID E / GENE
92	C628	ribosomal protein S17E
93 & 94	C665	malate dehydrogenase
95 & 96	C669	malate dehydrogenase
97	C694	ribosomal protein S3AE
98 & 99	C709	ADP/ATP translocase
100 & 101	C714	ADP-ribosylation factor 1
102	C721	calponin
103 & 104	C726	ribosomal protein L11
105	C736	nematode specific
106 & 107	C773	troponin
108	C834	nematode specific
109	C860	bystin
110 & 111	C863	troponin
112 & 113	C883	translation elongation factor eEF-1A
116	C888	40S ribosomal protein S16
117	C898	glyceraldehyde 3-phosphate dehydrogenase
118 & 119	C935	peptidyl-glycine alpha-amidating monooxygenase
120 & 121	C937	calponin
122 & 123	C942	peptidyl-glycine alpha-amidating monooxygenase

SEQ ID NO:	APPENDIX 1 (cont.) INTERNAL IDENTIFIER	FUNCTION OF POLYNUCLEOTID E / GENE
124	C954	arginine kinase
125, 126, 127	C969	calponin
128 & 129	7235653	ribosomal protein L18A
130	8005381	neuroendocrine protein
131	7235496	pyrroline-5-carboxyla te reductase
132 & 133	7275710	protein phosphatase pp1-beta catalytic subunit
134	7923685	nematode specific
135	7641370	40S ribosomal protein S11
136 & 137	7923404	nematode specific
138	7797811	ATP-dependent RNA helicase
139	7143613	predicted phospholipase D

Appendix 2:

Exemplary genes used for RNAi vectors.

Promoters:

Constitutive:

Super Ubiquitin from Pine
 CCCGGGAAACCCCT CACAAATACATA AAAAAAAATTCTT TATTTAATTATC AAACCTCCACT ACCTT
 TCCCAACCGTTA CAATCCTGAATG TTGGAAAAAAACT AACTACATTGAT ATAAAAAAACTA CAITA
 CTT CCTAAATCATAT CAAAATGTATA AATATATCCACT CAAAGGAGTCTA GAAGATCCACTT GGACA
 AATTGCCATAGTTG GAAAGATGTCA CCAAGTCACAA GATTTATCAATG GAAAATCCATC TACCA
 AACTTACTTCAGAA AAAATCCAAGGAT TATAGAGTAAAA AATCTATGTATT ATTAAGTCAAAA AGAAA
 ACCAAAAGTGAACAAA TATTTGATGTACA AGTTTGAGAGGA TAAGACATTGGA ATCGTCTAACCA GGAGG
 CGGAGGAATTCCCTA GACAGTTAAAG TGGCCGGAATCC CGGTAAAAAAGA TTAAAATTTTT TGTAG
 AGGGAGTCCTGAT CAAAGTTTAT GATGGAAATAGA TTCAAGCACCCTC AAAAACATTCAAG GACAC
 CTAATTTGAGT TTAACAAAAATA ACTTGGATCTAC AAAAATCCGTAT CGGATTTCTCT AAATA
 TAATGAAATTCTCA TAACCTTCAGG CAACTCCTCCCC TAACCGTAAAC TTTCTACTCTC ACAGGT
 TAATTAATCCCTTA AGAGTAGATAAA GAAATAAGTAA ATAAAAGTATTCT ACACAAACCAAA TTAT
 TTCTTTTATTACTT AAAAAACAAAAGTTTATTAT TTACTTAAATGG CATAATGACATA TCGGA
 GATCCCTCGAACGAG AATCTTTATCT CCCTGGTTTGTAT ATAAAAAAAGTAA TTATTTGTTGGGG TCCAC
 CGGGAGTTGGATCTC TACAGACGGCT TTACATACGTCT CGAGAAGCGTGA CGGATGTGCGAC CGGAT
 GACCCGTATAACCC ACCGACACAGCC AGCGCACAGTAT ACACGTGTCAIT TCTCTATTGGAA AATGT
 CGTTGTTATCCCCGGCTGGTACGCC ACCGATGGTGAAGGTGTCGTGTT GTCGTGTGCGGT AGCGG
 GAGAAGGGTCTCATC CAACGCTATTAA ATACTCGCTTC ACCGCGTTACTT CTCACTTTCTC TTGTC
 GTGTATAATCAGTG CGTATTTCTCAG AGAGCTTTCTCAT TCAACCCGGG

Strawberry Banding Vein Virus 1
aagcttttcactgtgggtaattcattaatctatccagggtaaaaacctaaggaga
tctctttctccaaaagacctctacagggcaatcaaaaactacagaaccagagttt
gtagtgcacagatgaccaatctacactgagaatcacgagtaccttcctagagtggg
aaaatgatgacatcatttcataccactggattgaggtaggactatccaatggaa
aaattccatgggacaagtcatataagaagaccgcaacagtgcagttatcttcagaga
taactgcactcagacctaaggataaaagcagttatataatcagtgtactaagatct
tcgcagattcaaqaagaagctt

Nematode Inducible:

Trypsin Inhibitor from *Arabidopsis* (clone#6598343)

Arabidopsis Transmembrane Protein from Arabidopsis
(clone#6468048)

cccgaaaaattggcaacttcttctgtgggttccaaaagaacgaatcaatatgtgc
aacaagaagagctccagaagcagtcatcttctaaaatctaatctaacaacagctca
agaagaaaaaaattccatagctagagagaacacaaaagt cacaagacgacgtcgtaga
ggcacaaaagtcaaacctgaatggcttaagccgaactgagtggtttgcactagaccat
catcagaaaaagtctccaagacggtagtcggatgttagatcgctcaagtaattttgg
tttgggtctcacgtttcagctgccatggattcagttggctttccta
tctctaaaggcccaattcatttaggttagttattgatcattatccttactata
aaggctcgccttcgagaaatttaggttctgtctgtctcgtcactcagggtt
tgtgcctcaacgactgctcacttctagcttgcattttcttcgttataatgtat
actgtacatttagattattctgtttctcgagctctgtctatagatttgcattttt
tttgggtcttgcaggatcagatcttagctaaatttagagacaagctc
aaaatgaggtacttgacgcatttacattcactgtttaaatttagagaacaatacgt
ctctgaatcgtgattcagagacgtattgttcttctgtcatatgcaataagtttatt
agagaacaatacgtctgaatcgtgattgtttttggatgtgcgttattgatacgct

ttatgtatgttaatagtctaggattgacacgaaaggttgttcgtcagtttcgcataaaat
ctctttactaaggcctctaaatttggatgacaatctaaatcttgcctcataaaaat
ttagggttattaagataagattattttgtatggtagtgtctataatgtgggttgc
atgttgagggttgcataatgttgttattttgtttagttattgtcttaactct
gttcttgcgggtaatacagtaagcgtttaggtgaggccgttcgtgaagccatcac
tactatcacaggaaatccgaggcaaagaaacgtaactttgtcgagactattgagct
ccagatcggtctgaagaactatgaccctaaaaggacaagcgtttagtgatctgt
caagttaccacatatccccgtctaaaatgaagatctgcattgtcgagatgccc
gcatgttgaagagggtgatatatctttcatggaaaattgtatctttgtctgttt
cttgcataatgggttgcatttcatttggctctattgttcatgttgc
ttgtatatgtcttcgaatgttagatgcattgtttcgaaattgttgcattgttgc
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caagaacaagaaactcgtaagaagcttgcatttttttttttttttttttttt
tgagtctgtcatttaagcagattcctgttttttttttttttttttttttttt
caagttctggctacagctaataattccattgttgcatttttttttttttttt
gataggttttagtagtctatttttttttttttttttttttttttttttttttt
tttatcctgtgagattatgttttttttttttttttttttttttttttttttt
ctttacacaggaaaattcccaactcttgtgagccaccaggaaatcctggagtcaaag
gtgaatgaaacaaaaggcaacagtgaagttccagctgaagaagggttgcattgg
gttgcagttggtaaccttccccggg

Diaminopimelate Decarboxylase from *Arabidopsis*
(clone#4159709)

cccggtggcaactgagatataagagggaaagggtatttcatgcaattttttt
tatttttttgaatgaatgcaaaatttattcaaaaaaaaaacctggctacatc
aagtacttcatttctgagttttgaaaaatctaaagacaacaaagacttacaatt
taataaaaaataaaaaataactttatcactctcaacgaaattgtgatthaataa
cgtatctcttggtaaaacagcgtttatttgcacgaaattgttataaatgaataaaat
gataatagaaactagtgtgtacgtaaaataccctctcatttgcacaaataacggta
tgtatcatgagtattgcatacgcacagcgtcttaatagtgtgcatttcaggagaaaa
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taaaaaaccaatgtcatttcttaatgttattcaaggtcatttaataaaaattgtacac
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ttgatatagttacccaaaagtgatataattcccttataatattttcttattttct
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aagttttctgtttgttttatatctatagccaaagaaattttctcagattacaaga
agttactgagaaaaacaaaaaaaaacttatgaagcatgaaagactaattaacgag
gtgattaatttgcataatttgcataatttgcataatttgcataatttgcataatttgc
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acagattgtgaatgagaccaatcaatggtcataaaccgggtttaaccggca
agtcatcttggcataattccattcgttattcattcatgcaagaccctctgatataaa
ccaaagactccattacaatattcttcgatcagcagacttatttcaatgtgt
tacctcttcgtgactcttgtgtgtggtaaaggctactcgagatgtgtcggttat
atatagcatacatatacaaatgcacaaaataagtatatttgcataatttgcataatttgc
atattccatttctatatgcattggctgggattttgcacaaaaccctaattcaagaat
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aagtaagacaaagaaatacagatataagatggctgtagaaaccaggtagaggaatttgc
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gaaatggaaatagtgcattataattcatcagattctatataatgcattttgcataatttgc
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tgtggttttatttttgttagctagaaacttgcattttttgcacactttgcacaaatttgcataatt
tgtggttttatttttgttagctagaaacttgcattttttgcacactttgcacaaatttgcataatt

tgctttgatgatggtccggcccgaa

Peroxidase from *Arabidopsis* (clone#4006885)

ccccggggcaatcaaggtaacgaaggaggatcagcggaaaaggatgggctatatttgaggat
tttttctgcgtgtaaatgcgttgcattccatgcggacatataactgaaga
ataaaactcaactcattgtttctgggtgtttctgtatcagattcctcgatgc
ctgcacatttctgtgtggggcttatttataaaaacaagagtagagcgtgtggtaa
tcttcataatcttctacaattccactccattctaatatttctcacgtgatata
acacacactcaatcactgtgtactcgtatggatcagcgtggaaactgtatgcattgc
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tttagttaatcttaggtttatattgtttattacttcttgcatttgc
ttcttatttgcatttgcatttgcatttgcatttgcatttgcatttgcatttgc
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atattatcatgttgcatttgcatttgcatttgcatttgcatttgcatttgc
aaaaataattactttagtgcatttgcatttgcatttgcatttgcatttgc
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tcttgcatttgcatttgcatttgcatttgcatttgcatttgcatttgcatttgc
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ttcttgcatttgcatttgcatttgcatttgcatttgcatttgcatttgcatttgc
tcatcccaatttagtgcatttgcatttgcatttgcatttgcatttgcatttgc
aaatacgttgcatttgcatttgcatttgcatttgcatttgcatttgcatttgc
cctcatccccgggg

Mitochondrial Uncoupler from *Arabidopsis*

(clone#4220510)

cccggtatgtgtgatgtgaaaggagaagaagaggaaacaaaggatattatgttagc
gagttttgtttgtacgcgggtttgtctgttcaatgttgcgaaacgcgatgaga
gagtgtctgattattaaagaaaaccctaattaagtcaagccgcgggtataaaaat
agtcaaaaagtagaaaaacgcgtgtgagtgagacagagacagccattgttgc
ttatggcattataagcgagacgtttaattggctttccattggccgaaaaca
aaagaaaacgtcgccctgagagattcgaaactctgcggcagagccatgtacttagca
ggcacacgccttaaccactcgccaaagcgactgttgcattgagttagacaaaatc
attaaaattctctattatgattctcatagtggtgttatattgtggatctactaa
aaattcttgcattattactttattttgtgaatttagttgatataaggtaagtacaa
agtttaactttattttactcaaaatttacatgagattactgtatttatattgtttcc
tttggtatatagacgtactatagttttagaaaaaccataagattccattatattc
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aggactttttttttgtgaagacgaggaggaggactttggatccagtc
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aatcttcattcaaccgaaccaaccaaaagtcttccaaataatattcaagcaccatc
cttggaaactcatacatactacagtctacactcttcatttcttcaacgctca
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63

tcaaattctgggttgcattttgtactatccggaaacatctcaatgtcccccaaa
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aaaagtataatataactgcctctaattttcgcccttctaccgaagaatctccact
cttgcctcttgcggaaaccctaaaccagaagcaccagattttcaacttttcca
gagaacaatagaaaaaccctaaacttgcgttgcgttgcgttgcgttgcgttgcgttgcgtt
tttggatttcttgggtcatcattttggagcttaccaccagcgaaaaattataaa
cttccatcgattcctggcttcctctcgctctgtcatgtgcataatcgccgg
actgatcctcactgtcacctctgttcccggg

Stress protein from *Arabidopsis* (clone #6598614)

Pectinacetyl esterase from *Arabidopsis*

(clone#6671954)

ccccgggtggtgtggggacaatggatccggctcgctagcaacaqqctaaaaagatta

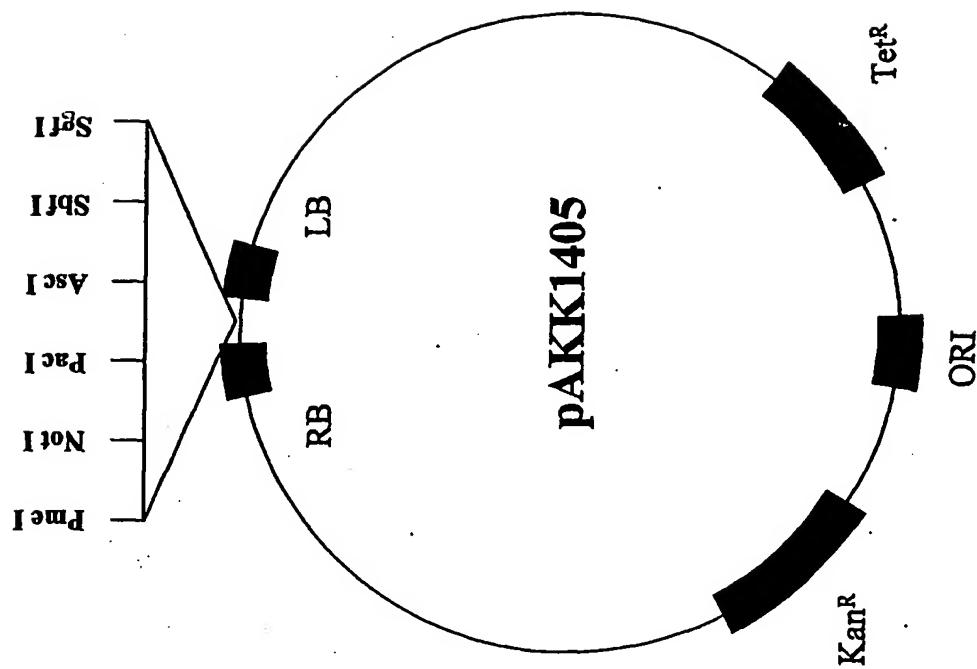


FIG. 1

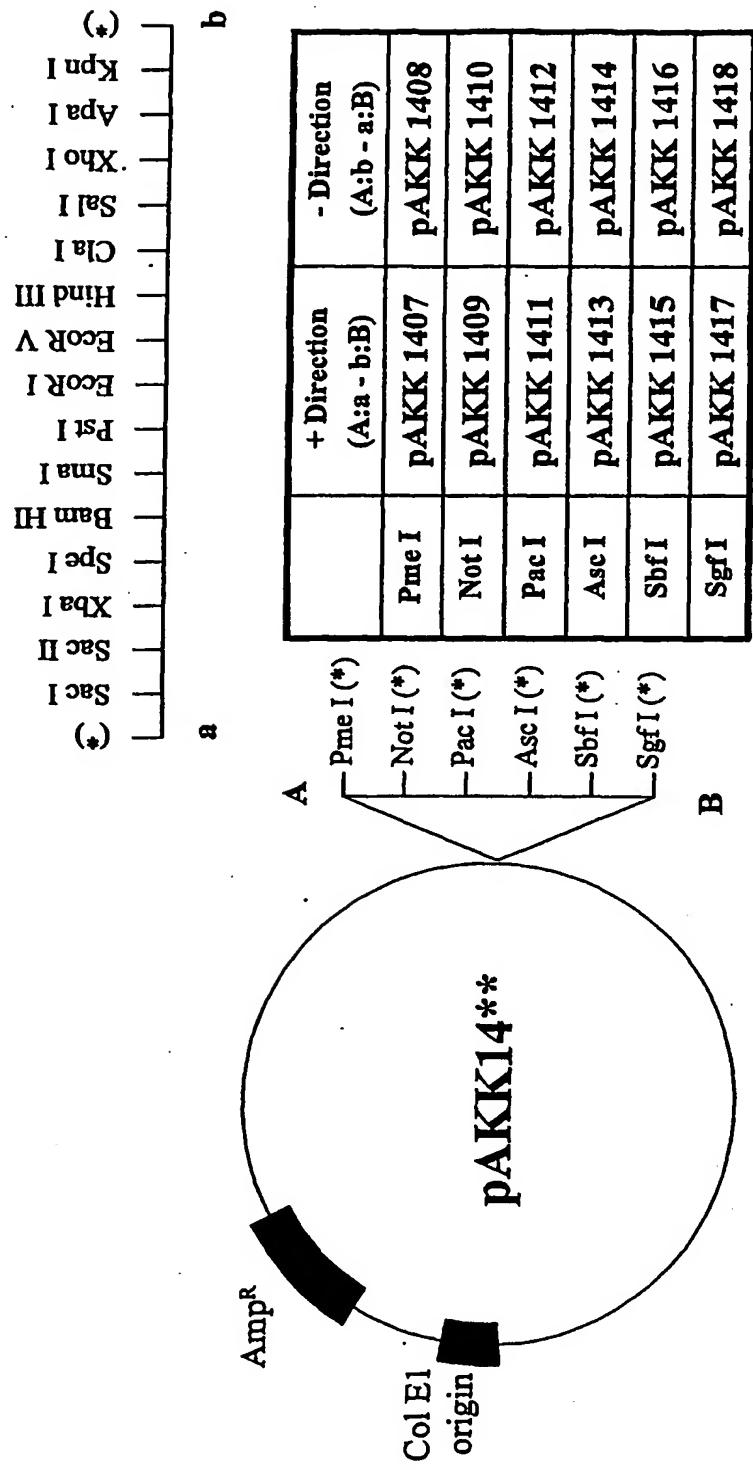


FIG. 2

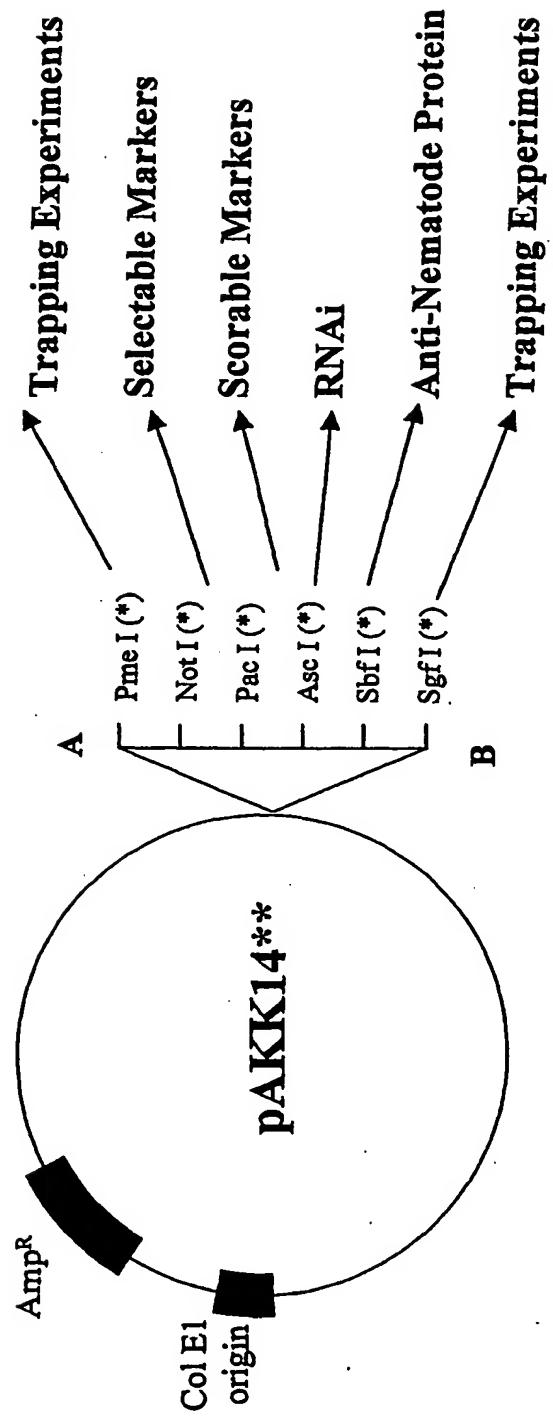


FIG. 3

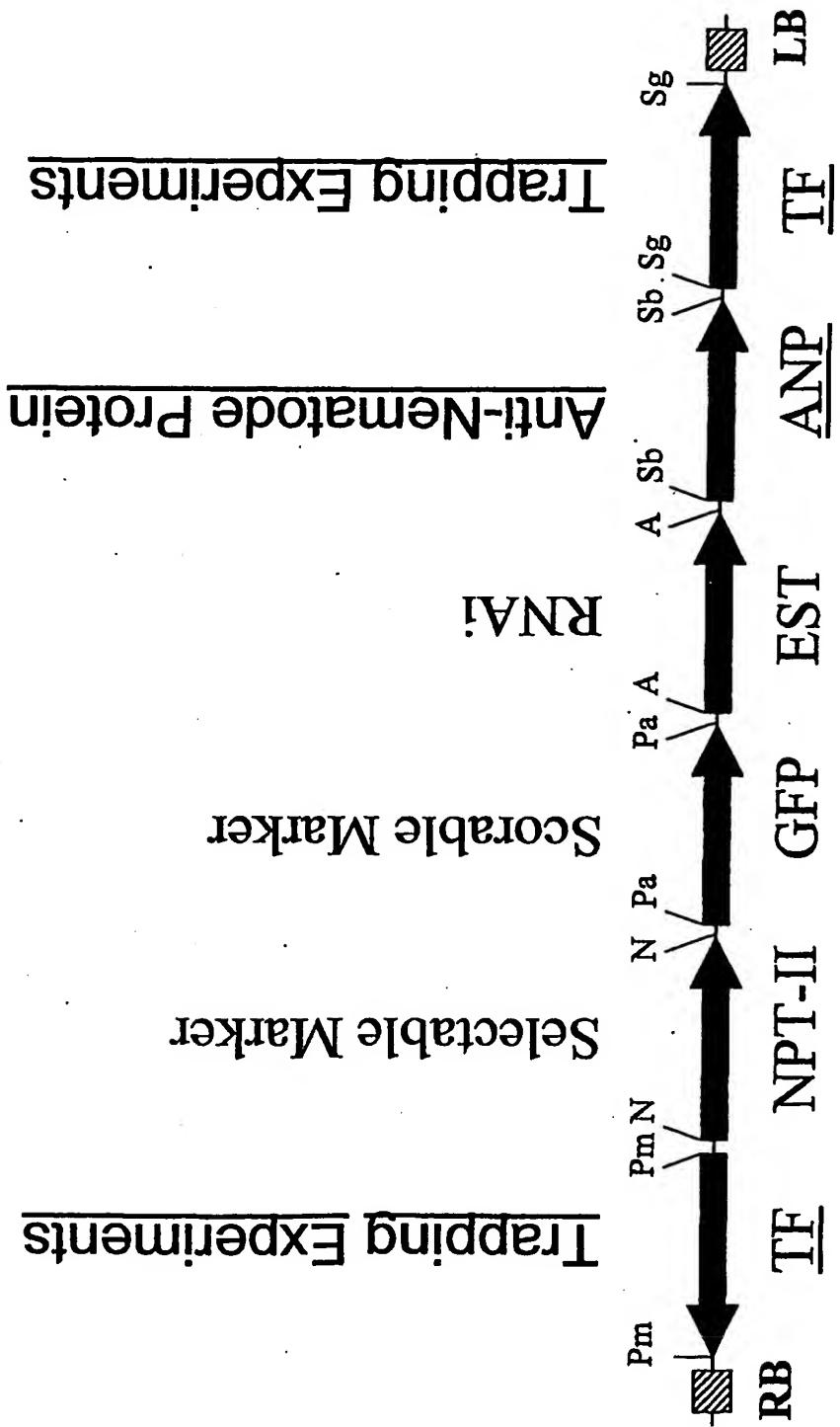


FIG. 4

Selectable Markers

pNOS / NPT-II / tNOS

pSU / Bar / tNOS

pSU/ Intron / Bar / tNOS

pUBQ3 / Intron / PMI / tNOS

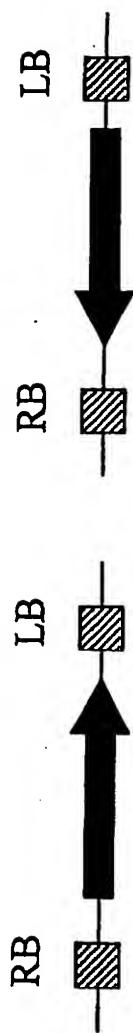
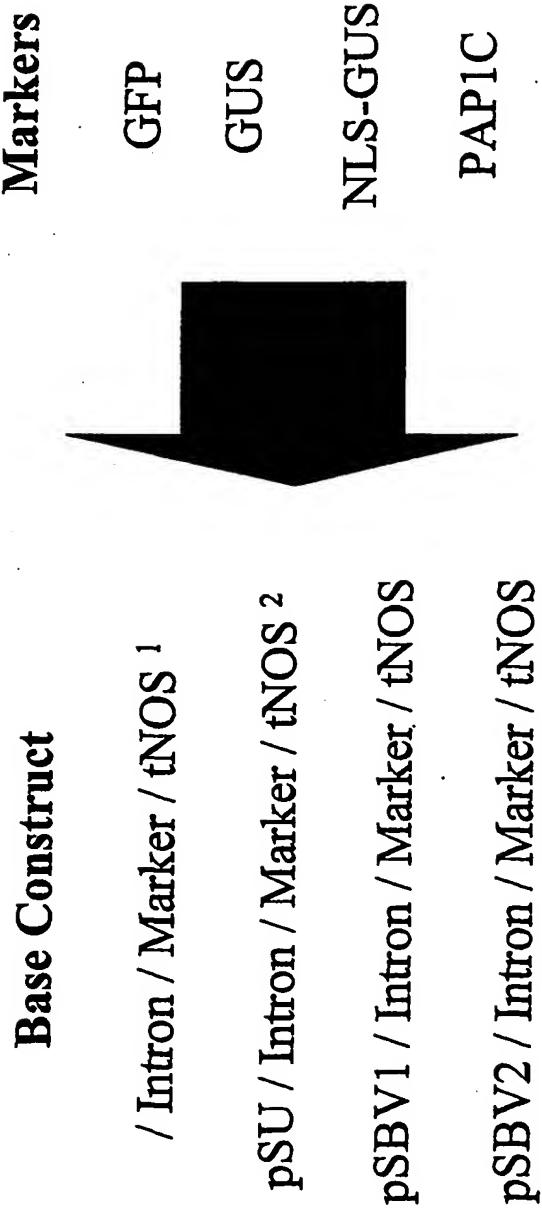


FIG. 5

Scorable Markers



- 1 Construct useful for promoter analysis.
- 2 Construct useful for high constitutive expression of genes of interest.

FIG. 6

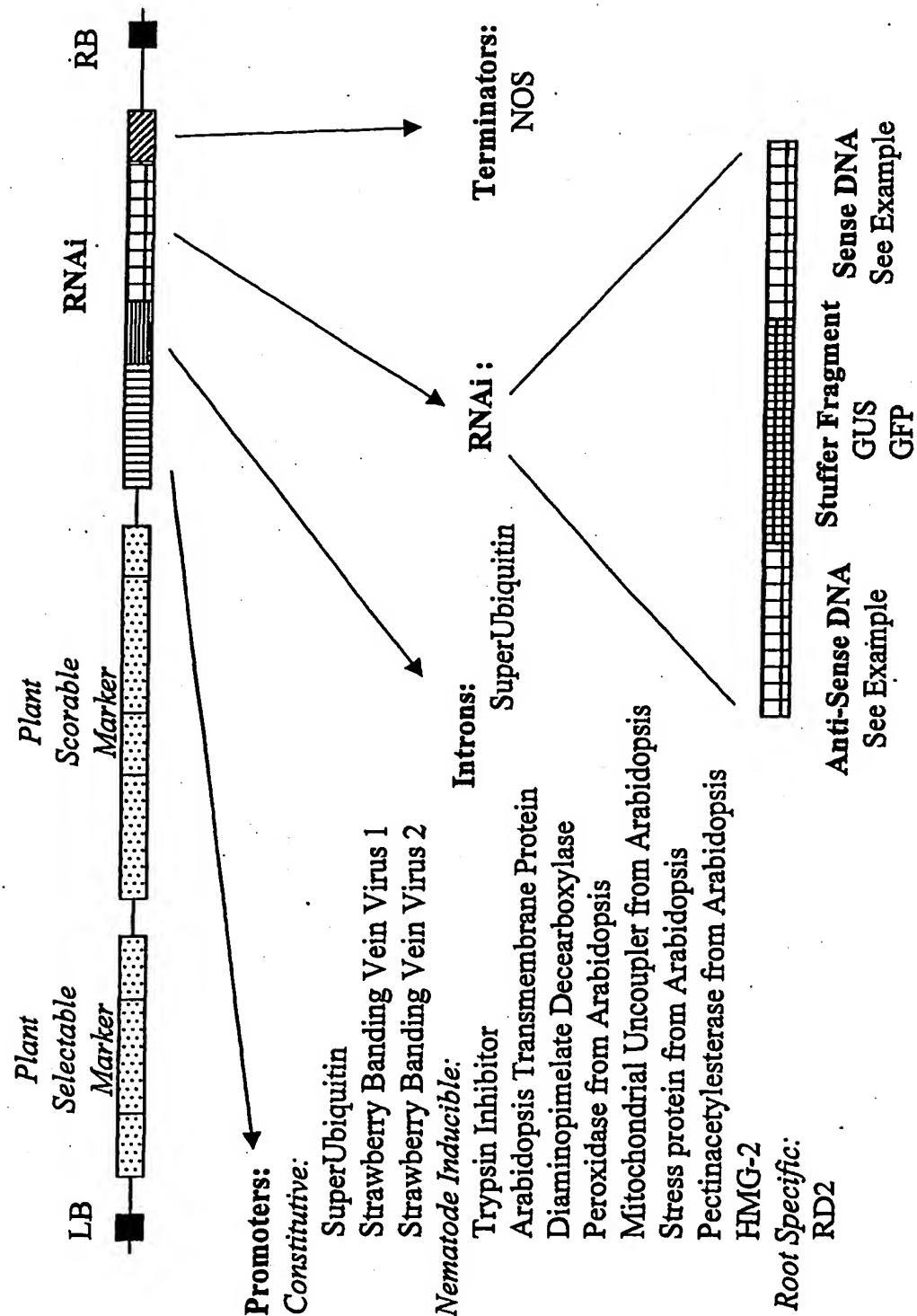
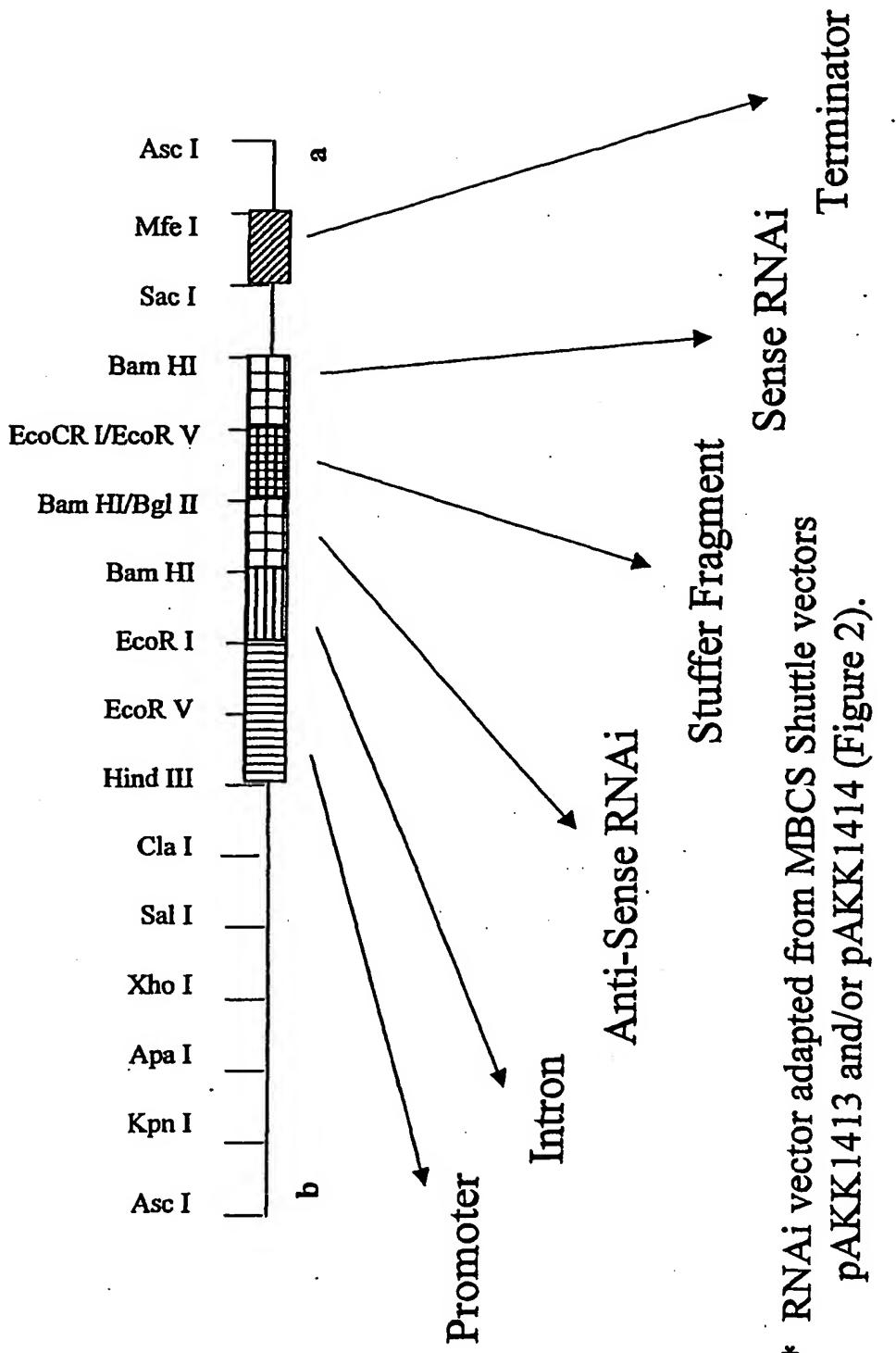


FIG. 7



* RNAi vector adapted from MBGS Shuttle vectors
pAKK1413 and/or pAKK1414 (Figure 2).

FIG. 8

AKK110P1
SEQUENCE LISTING

<110> Mushegian, Arcady R.
Taylor, Christopher G.
Feitelson, Gerald S.
Eroshkin, Alexey M.

<120> Materials and Methods for RNAi Control of Nematodes

<130> AKK-110P

<140>
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<160> 139

<170> PatentIn Ver. 2.1

<210> 1
<211> 165
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gtttggcggtt gcgcgctgctg gttgagaagg acaccgttca ggtgg 165

<210> 2
<211> 342
<212> DNA
<213> *Globodera rostochiensis*

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ttcgacaagc gccggcaatt tggctgttga gaaagagggg aaggccacgc acaccatcaa 120
ggtgttcaac ctcaaggacc cggccgagat caaatggct gaggtggcg cggaaatatgt 180
gatcgagtcc accgggggtgt tcactaccat tgagaaggct tcggcacact tgaaggggg 240
cgccagaag gtggtcatct ctgctccgtc cgctgtatca ccgtatgtacg tgatggcg 300
caacgaggac aaatatgacc cggccaagga caacgtgatt ag 342

<210> 3
<211> 205
<212> DNA
<213> *Globodera rostochiensis*

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gaagggcatt ttgggttaca cagaggacca ggtgggttcc acggactttc ttggagacag 120
tcgctcgatcg atttcgacg ctggggcggtg catctcgatgg aaccgcact ttgtcaagtt 180
ggtcagctgg tacgacaatg aattt 205

<210> 4
<211> 167
<212> DNA
<213> *Globodera rostochiensis*

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tcgtccatccgttgcattt gtcaattgtg gcccattaaaga gggccgtttg ggttagttt ttgggtttcc 120
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<210> 5

AKK110P1

<211> 41
 <212> DNA
 <213> *Globodera rostochiensis*

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41

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 <212> DNA
 <213> *Globodera rostochiensis*

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 cttaacgcct ccacgacgg 79

<210> 7
 <211> 168
 <212> DNA
 <213> *Globodera rostochiensis*

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 ctttccgagt cttttccgc ctttccgcg tccggacatt ttgttgtttaa atcagaagag 120
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<210> 8
 <211> 330
 <212> DNA
 <213> *Globodera rostochiensis*

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 gcccataaaag aaagacgaag aggtattggc taagaacacg cccacatttgc tcgtcggaa 180
 gcccggacgt cttttggct taggacgcac tggacatctg aagctgaaag gcgtcaaattc 240
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<210> 9
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 aaggaaaatg agaaga 136

<210> 10
 <211> 141
 <212> DNA
 <213> *Globodera rostochiensis*

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 ttgttgttca tcactttctt cagcagcgcac aatacggcca atccggtaa agggccaaag 120
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AKK110P1

<213> Globodera rostochiensis

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 aagtagccgt atttgcgaaa t 141

<210> 12

<211> 37

<212> DNA

<213> Globodera rostochiensis

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<210> 13

<211> 161

<212> DNA

<213> Globodera rostochiensis

<400> 13

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 ctttggtgca aatggcaaaa cgccaaaaat aatggtcgaa gccgtacaca accgcccacg 120
 ccacagcgc aaccccacac caaatgcgaa atttacgcgaa a 161

<210> 14

<211> 306

<212> DNA

<213> Globodera rostochiensis

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 agtttgcata ctcgattgtt agaagttcgg ggtttagac cggaaaaac agtgc当地 120
 gacaatctg agatacgcac ttgtgcattt aaaaacacgtg aaattttgtt gtcgcagcca 180
 atcttgggg agctcgaggc acctttaaaa atttgggtgt acatcgcgg acaatataat 240
 gatcttctga gattgttcga atatggggg tttccaccgg aagcgaacta tctatttctt 300
 gggac 306

<210> 15

<211> 261

<212> DNA

<213> Globodera rostochiensis

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 tgaatgc当地 cggagggtcc tcaatcaatgttggaaagac cttcaactgac tgcttcaact 180
 gtcgtccat tgccgctta atcgacgaaa agatcttttg ctgcacggg ggctgtctcc 240
 tgatttgcata aacatggcag c 261

<210> 16

<211> 151

<212> DNA

<213> Globodera rostochiensis

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 accaaaaaag catttgc当地 gacttgcgc accaaaaaaa tggatgttgg acaaatttggg 120
 tggcgaaaa gcccacgtc cattgtgcgg a 151

<210> 17

<211> 306

AKK110P1

<212> DNA

<213> Globodera rostochiensis

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ttgagaagac	aaacgaaacg	tttcgtctgg	tgtacgtgt	gaaggggccgt	tttgcatcc	180
atcgaattca	aaagctggag	ggccagtaca	agctgtcaa	agtgaagaag	caggccgtcg	240
gggacaagca	ggtccctac	attgtcacac	atgacgcgcg	caccattcgc	taccggaccg	300
ctcatc						306

<210> 18

<211> 528

<212> DNA

<213> Globodera rostochiensis

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ggagcaagca	gacgacctt	cggttggct	ttgttcgtcc	attgggttgg	agcatcgccc	120
gttcctaccg	tatacaaacg	ctgtataaaa	tgaaacaatt	cgatttagtca	atttgatccc	180
gttcaatctt	agccatttgg	cgcttgaaga	tatgcaaatt	ggcaatttta	ttgtgaagcg	240
tgggacacca	attgtaccgc	aggtcagcag	tgttctgttc	gacgaaaaac	tgtatccgga	300
gcccgatcg	tttttgcgg	aacgctttct	ggacgtatgg	ggccgttga	agaaaagcga	360
cgaacttatt	gcatttgggg	ttggggaaaag	gcaatgtgcc	ggcgaagctt	tggcccgaaat	420
gacactttt	ctgtttgcgg	ctaatttctt	tctcgctac	aaagtctcc	cgtccgatcc	480
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<210> 19

<211> 335

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<213> Globodera rostochiensis

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ctccggccca	ttgggcgttg	gccataaaatt	tatgagccgc	ggtgcgggtg	agggcgtcca	240
acagctaggc	cccgaggggc	ccttgagca	gcccacacag	gtgaagagt	acaatgttct	300
cccccgat	tgcgagcctc	caaattccctg	tccga			335

<210> 20

<211> 52

<212> DNA

<213> Globodera rostochiensis

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ggacggctgc acggaacagt tcgagaacac tgccgagtt tcgcgcagct ac

52

<210> 21

<211> 190

<212> DNA

<213> Globodera rostochiensis

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agcaggatct	ggagcaattt	ctggccaaca	acggactgca	caaatcaatg	attgccaaga	120
aattccatct	cacgcggcg	gaggagccgc	gccgtcgaaa	acgctttgt	cggccggctt	180
cggccaaccg						190

<210> 22

<211> 52

<212> DNA

<213> Globodera rostochiensis

AKK110P1

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 <210> 24
 <211> 77
 <212> DNA
 <213> *Globodera rostochiensis*

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 aacagaccgg aacagca 77

 <210> 25
 <211> 439
 <212> DNA
 <213> *Globodera rostochiensis*

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 aacaacgtca gcagcaacat gtttggtc aacaacagca gcaacaacag aatttccaa 180
 aaccggccgc cctatcgat actcacagcc accaacaaca aaaacaacca ccacaagcgt 240
 cacagtgcgt gttgtcaatg aaaagtggca atggtgtcgt tgggttccg caacaatcgc 300
 agcagcacca ctaccaacag cggacactga cgccactgaa gcacacatcc gcatcctca 360
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 gcgccacggc cactgatga 439

 <210> 26
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 <212> DNA
 <213> *Globodera rostochiensis*

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 aacaaaaact gtgaagaagg cgtcgcgcgt cattatttgg aagtattaca ccaaattggg 120
 cctcgacttt cacaccaaca agcgcattt cgaggagggt gccattatcc caagcaaaacg 180
 gatcggaac cgaattgcgg gatttacac acatctgtg aagcgcattt agctgggccc 240
 tggccgtggc atttccatca aattgcagga ggaggagcgc gacgtcgcgc acaattacat 300
 gcccggaaatc tcttacctgg atgcgcagaa tcaccagatg atcagcaccg accaagagac 360
 gaaggatatg gcgaaatcc tggggctagg cctcaactt gaaatgaaaag ggcctttgac 420
 gagttggcgcg gctggcgcag gacgtcgtt agtgcggaca attgcattt ttgttggaaa 480
 atcatcgatg ttttggatga taatgcgcgtg ataaatttt gttgatttt 539

 <210> 27
 <211> 179
 <212> DNA
 <213> *Globodera rostochiensis*

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 cggccgaaaa gctgtcgccgca gaaaagatata atgatgcccc gaagcgaaaa gcacagcgcac 120
 ttaaggcaggc caaacaagaa gcccaggcgg agatcgagca gatcgncag gagagggag 179

AKK110P1

<210> 28

<211> 133

<212> DNA

<213> Globodera rostochiensis

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gcaaaaattat ttgggcacgc gcgacgacat cgagcagcaa ataaagcgcg agacagaaga 60
 gtcgcgtggag gcaatgaatc gcaatgtcgc ggcgaacaaa cagcaggtca ttgtacgtct 120
 gctgcagttg gtg 133

<210> 29

<211> 482

<212> DNA

<213> Globodera rostochiensis

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 caaaaaggcga tgtgttttgcg aaagatcgc caattgttct cgttctccctc gacattccac 180
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 ttggccgtgt ggaggctgtt accacggaaag agcaggccctt caaggacattt gactacgctt 300
 ttcttgcgg agcgatgccc cgaagagagg gaatggaaacg aaaggacctt ttggcggcaa 360
 atgtcaaaat ttcaagtcc caaggcgaag cattggcccg cttttccaag cccgtncgtc 420
 aaagttctcg tggtgggcaa cccggccaac acgaacgcgt acatttgcgc aaaatatgcc 480
 gg 482

<210> 30

<211> 605

<212> DNA

<213> Globodera rostochiensis

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 ttagacggcc tcgttttca ttttttgtt atgcgtcac atcaggcga gctgccagtg 540
 atttggcacc agacactgtt ggctttgtc gagcgttacg caaaaagacat aagtgcagaa 600
 cagag 605

<210> 31

<211> 112

<212> DNA

<213> Globodera rostochiensis

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 aatgaggaaa gtgaagcaaa tgtgcccgtt tatgcgcgtt atgatgaaat gg 112

<210> 32

<211> 105

<212> DNA

<213> Globodera rostochiensis

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<210> 33

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<211> 425
 <212> DNA
 <213> *Globodera rostochiensis*

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 tacgctcctg acgctgaggc ttacacctt ttcacggcgt tggtcgacc gatcatcaac 180
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 aaaacgcana tgctgaccgg atctcgaccc cgaggggaaa atttatcaat ttgcacacgc 300
 gttcggttgcg gccgttcct ttaagggata cccgggtcaa cccgtgctt acnaaaggan 360
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<210> 34

<211> 581

<212> DNA

<213> *Globodera rostochiensis*

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 tccttgacgg tccaaagccg cggtcagttc cgtgcccgtgt tttttaaaag aggcggagag 120
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<210> 35

<211> 102

<212> DNA

<213> *Globodera rostochiensis*

<400> 35

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 cccatcaaag catccggaga aacattaagg aagtttatttgc 102

<210> 36

<211> 34

<212> DNA

<213> *Globodera rostochiensis*

<400> 36

tgcaaatgt gcaaaccacca cgttcacaa gatg

34

<210> 37

<211> 100

<212> DNA

<213> *Globodera rostochiensis*

<400> 37

tcatgttgcg cccaaatctc gcttctggta ctttacgagc atgctgcgtc gagttaaaggaa 60
 aacacacggaa gagatcggtt cgtgtcaaga ggttttcgag 100

<210> 38

<211> 176

<212> DNA

<213> *Globodera rostochiensis*

<400> 38

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 gcgagtatcg ctgatgttac cgaggccggt gccgtgaccc aatgtatcg cgacatggc 120
 gctcgtcacc gcgctcaggc ggatcgaatt caaatcatca aagtgcaaac ctcaag 176

<210> 39
 <211> 155
 <212> DNA
 <213> Globodera rostochiensis

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 agcgcgcgtt ccaaaaacaa ccgatcggtt ttctgaacga caagttcaga acgcaaggaa 120
 ttgggaagaa ggcatccaaac aaggaccgtt actgg 155

<210> 40
 <211> 35
 <212> DNA
 <213> Globodera rostochiensis

<400> 40
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<210> 41
 <211> 70
 <212> DNA
 <213> Globodera rostochiensis

<400> 41
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 gcggacgatt 70

<210> 42
 <211> 85
 <212> DNA
 <213> Globodera rostochiensis

<400> 42
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 cgtgcttccg agatgtctct ctcgg 85

<210> 43
 <211> 193
 <212> DNA
 <213> Globodera rostochiensis

<400> 43
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 tggaaaagct cga 193

<210> 44
 <211> 219
 <212> DNA
 <213> Globodera rostochiensis

<400> 44
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 gaagacgtcc ggcgcgttgt tatcgctata ttaagaacaa gccgtatccg aagtgcgcgt 120
 ttgtcgccgg tgcgtccgac ccaaaaattc gcatttttga tttgggttaga aagcgccca 180
 ccgttgcgtca attcccatgc tgcgtgcata tgatatcg 219

AKK110P1

<210> 45

<211> 489

<212> DNA

<213> *Globodera rostochiensis*

<400> 45

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atgttgcct	gcgctggcgc	ggaccgtctg	cagactggga	tgcgtggcgc	gttccggaaag	180
cctcaggagc	tgcgtggcgc	tgtcagcatc	ggtgatatgc	tgtatgcgt	gcgtattcgt	240
gaccacaccc	aagctcaccgc	attggaggcg	ttccgtcggt	ctaaattcaa	gttccctgtt	300
cgtcaataca	tcgtcttgc	ccgcaagtgg	ggcttcacca	aattcgatcg	cgaggtatac	360
gagaaatacc	gcaaggaggg	ccgtgttac	cctgacgggt	tgcattgca	gttactcaag	420
caacacggac	ccgctgaagg	agtggctcaa	gaacccatt	taatcttctg	tttgcgttgc	480
gactcttgg						489

<210> 46

<211> 101

<212> DNA

<213> *Globodera rostochiensis*

<400> 46

gaattcccccg	gctcgagccg	ggttgacgt	gtccctcctcc	acccctctc	actgcgttcc	60
gtccctccttc	agccggaaat	tgttcctgt	gctgttgcg	g		101

<210> 47

<211> 485

<212> DNA

<213> *Globodera rostochiensis*

<400> 47

tccaccaaag	tccattcgct	gtcgccagtc	catttattcc	acaaaaagat	gattccgtcg	60
tcgttccgt	gacgtcgtt	ggccaaacgt	tgccccgtc	accgtttca	ctgggtccaa	120
acccggcgt	ttattttgc	ttcccaaaaa	acttgcgtt	ggagcggccc	tgcacgagc	180
aaaacgacgg	ctccgaggag	gaattagccg	aagaagcgt	ggaaacgaag	gcgaagaggg	240
cgcacacgtt	cgtccgtt	ggccaaaggg	cgcaacacat	tgtccgtt	ggaaagcgt	300
cacaacacatt	tgtacgcctc	ggaaggaca	cgcaaaaggca	attcgatgg	aaaatgcaaa	360
gtgaacagca	acagaaaaag	gcttaaagca	aacggccgcg	actttctt	taatgaatgc	420
gcgcaccacgg	catgacaatt	ctttgtgt	atgtgttgc	atrtttatga	tcggtaaatg	480
taaca						485

<210> 48

<211> 651

<212> DNA

<213> *Globodera rostochiensis*

<400> 48

atctttcaa	gggactgttc	ggcaagaagg	aatgcgcatt	tctgtatggtt	gggttggacg	60
ctgtggaaa	gacgaccatt	ctgtacaatg	taaagctcgg	cgaaaattgtc	accaccatcc	120
caacaattgg	cttcaacgt	gaaaccgtcg	aatacagaaa	catctcgatc	actgtttgg	180
acgtgggtgg	tcaagacaaa	attcgccac	tttggaggca	ctacttccag	aacacgcaag	240
gactgatctt	cgtcggtgg	agcaacgatc	qcgagcgtgt	gggcgaggcg	cgtgaagagt	300
tgtatgcgt	gctggcgagg	gacgaggatc	gacgacgcgt	gttgcgttgc	tgcctaaca	360
aacaggattt	gccgaatgcg	atgaacgcgg	ccgaactgac	agacagactt	ggactgcaca	420
acttgcgttt	ccgcaattgg	tacatccagg	ccacctgcgc	gacttccggc	gacggactct	480
acgaggact	ggactggctg	agcaaccagc	tcaagaacag	aggctaagct	gggttgggt	540
ctgttgcact	tgcggcggtt	attgatgcgt	attgaattt	tttgcgttgc	tgcgcgcgca	600
gctttttgt	gggacgtccg	attaaattttt	ataattattt	tattccgtgt	t	651

<210> 49

<211> 660

<212> DNA

<213> *Globodera rostochiensis*

AKK110P1

<400> 49

gaattcccaa	gtttgagatc	aattcagttt	cacttagaca	aaaatgccgc	cgaaattcga	60
cccaactgag	atcaaataatcg	tgtacctgcg	ttgcgtcggt	ggtaaaattt	gtgcaacatc	120
tgcacttgc	ccaaaatgtt	gcccacttgg	attgtcgccc	aaaaaaattt	gtgaagacat	180
tgcgaaggcc	acacaggact	ggaaagggtt	taagggttacc	tgcaagctga	caattcagaa	240
tcgtgtcgcc	aagatcgacg	ttgtcccattc	ggccgcctct	ctgatcatca	aagagttgcg	300
cgaacctccg	cgagaccgca	aaaaagtcaa	aaacgtgaag	cacaatggca	acctgaccat	360
cgagcaagt	atcaacattt	cgcgtcagat	gcgcctctgt	tcaatcgcac	ggaagttgca	420
gggcaccgtg	aaggaaattt	tggaaccgc	ccagtcgtt	ggctgcacca	tcgatggaca	480
acatccgcac	gacattgtgg	acgcgtatcg	agggggagac	atcgaaatac	ccgaggaata	540
aagaaaggac	ggcgcctccg	attttgtgg	gacggacatt	ggaaatttga	ggtgaatgag	600
ttgccaattt	cattcattca	tcaattgttg	ttattgtntgg	tacggataaa	tttgttaattt	660

<210> 50

<211> 625

<212> DNA

<213> Globodera rostochiensis

<400> 50

gtgccggaaac	agacgctcga	ggagggttagc	cgtctgcagc	ggacgagctc	tttgttggac	60
gtggcaatcc	gggacggcgt	cccttacccc	ccactgcctc	ctacaaaccc	atccccccgaa	120
tacatgaaca	tgctgaccgg	ctcccttctcc	gtgccaattt	tccgcattctt	ctcgggcgcc	180
atcgaccgt	acagacccctt	gttggccgtg	tacacttaca	acacttacca	cgggtacttc	240
cccttacccgca	actaccgcgg	ctacacccctt	gcaatgttt	actgtacga	ccgataactat	300
tacttctcg	cgctgtacaa	acgaagcatg	ttcccccaccc	gcttccaaaca	ttgtgactat	360
aaagcgaacc	cgcaatttt	gcactaccgg	cacacccctt	gggactatcc	ctaccaggc	420
aaatgggtcg	actacgacaa	ccctcccaat	taccggccct	actacaacca	tcgccttaac	480
ggatatgtctc	ggccgtatca	ctaccgggtc	catgcgttgc	cccacccgtt	caattaccgg	540
gaaggaatgg	tcaggaaacg	ggtctgacaa	atcgaaactgc	tccaaatttga	cgttgtccgc	600
attcgaaaga	agacgaaaaaa	agctt				625

<210> 51

<211> 402

<212> DNA

<213> Globodera rostochiensis

<400> 51

gaattccaaag	tttgagcaac	attttgaaaa	tgaccgaagc	aaaaaaactt	cccgagggtc	60
cggaaacttt	gctcaaggcg	cgcaaaatca	gagctgcgc	aaaggccgca	aaagcaaaaga	120
acaaattttag	ttctatcaaa	aaagcacgg	ccaagaagg	ggaaatctt	aaaagagccg	180
agcagtattt	ggtggagttac	cgtcagaacg	aacgccaattt	gcttgcgtg	aaacgtgaat	240
cgaagaaagt	cgcaatttat	tatgtgccag	aagagccaa	actcgcctt	gtggtccgaa	300
tcaaaggcat	caataagatt	catccgcgtc	ctcgcaaggt	tctgcagctt	ctccgccttg	360
gtcagatcaa	caacggcggtt	ttcgtaaaagt	tgaacaaggc	ga		402

<210> 52

<211> 433

<212> DNA

<213> Globodera rostochiensis

<400> 52

ccgaccggta	catcgcttgg	ggttatccga	gtcagaagat	catccgtcag	ttggtctaca	60
aacgcggta	cgccaaagag	aaggggacgc	gcattccaaat	aacgataac	acattgttt	120
agcgcagttt	gggcaagcat	gacgttattt	gtgtggagga	tatgtatccat	cagatttgg	180
ccggtcggtac	cgcacttcaa	acaggtgacc	aacttccat	ggcccttcaa	gctgagcaac	240
ccggtggcg	ggttcaagaa	gaagtccaaat	cactttgtg	gaggggaggcg	attatggaaa	300
ccgcgaggac	caaataaca	aatttttgg	aagaatggtc	taatggagg	gaagcggana	360
aagaaaggaa	atttggcggt	ttttctgtt	ttgttttgc	gataaaattgt	taactccaaa	420
aaaaaaaaaa	aaa					433

<210> 53

<211> 768

<212> DNA

AKK110P1

<213> Globodera rostochiensis

<400> 53

gaattcgttt	gaggtaaaac	tttattagcg	tatTTaaaca	tgtccgaagg	aggagcggaa	60
aagagttagca	gcgggtccaa	gggggggttt	gatgtcaaga	aatttgcgt	cgatcttgcg	120
tccgggtgt	ctgcggcg	tgttccaaa	actgttgg	ctccattga	acgtgtcaaa	180
ctcttgcgt	aggtaaga	tgcttccgt	cacatcact	ccgacaaacg	ctacaaaggc	240
attattgacg	tgcttgcgg	tgtccgaaa	gagcagggt	ttctgtcact	gtggcgtgg	300
aacttggcca	acgttatccg	ttatTTccc	actcaagcgc	tgaacttcgc	cttcaaagac	360
acatcacaaac	gcatcttac	ggagggactg	gacaaaaaca	agcagttctg	gtcggtcttc	420
gtcatgatt	tggccttcgg	agggtcg	ggcgcacgt	cgctgaccc	tgtttatccg	480
ctgggacttt	gcccgtacgc	gttggcccg	tcgatgtccg	aaaagctgg	tcccgcgagt	540
tcaacggttt	ggcccaactgc	atcgcaaaaa	tcttcaagtc	ggacggtccc	atcggtctt	600
accgcggctt	cttcgtctcc	gtccaggggca	tcatcat	ccgcgcgc	tactttggat	660
gcttgcacac	cgcgaagatg	attttcgcgc	cggatggca	gcagatgaat	ttcttcctca	720
catgggccc	cgctcagg	gtcaccgtgt	cgtccgg	cctccct		768

<210> 54

<211> 338

<212> DNA

<213> Globodera rostochiensis

<400> 54

gaattccagc	agattaattt	gaatggctga	gaacatcgaa	gagattcttg	ccgaaatcg	60
cggctccaa	atttggagg	atcaacgc	tttcgacat	ttcgaccgc	gaaagaatgg	120
ttacattat	gccacccaa	ttggacaaat	tatgaacgc	atggagcagg	atttgacga	180
aaagaccctc	cggaaat	tccgc	cgacgcggac	ggttccggca	aactggagtt	240
cgacgagttc	tgcgcttgg	tgtacacgg	ggcaacact	gtggacaagg	acactctgc	300
aaaggagctg	aaggaggcat	tccgactt	tgacaagg			338

<210> 55

<211> 267

<212> DNA

<213> Globodera rostochiensis

<400> 55

gaaattgcgc	ccgatctcag	cgacaaggat	ttggaggcg	cggtcgacga	aattgacgag	60
gacggcagcg	ggaagatcg	atttgcagg	ttctgggag	tatggcgg	cgaaaccgac	120
tgagaaaaga	gc	ccaaatccaa	acggacccgt	cccatttac	ctccatccgt	180
ccgtcgatt	attatattt	ccagtgg	tttccat	aaattcggt	aaagtaaaat	240
aatttgcga	aaaaaaaaaa	aaaaaaa				267

<210> 56

<211> 597

<212> DNA

<213> Globodera rostochiensis

<400> 56

gaattcgct	gacacttcgc	atccggagta	cagccacgag	cagagcat	accagaccag	60
catccccat	cagatgggtt	cgaacaagta	cgcctcg	aaggcat	ccggctttgg	120
acagccccgt	tgggggggtc	ttgacccgtc	catctcg	cagaacc	atgcgcaagg	180
aatgggtcg	ctacagtccg	gtaccaaccc	gttgcctt	caggcggca	tgaccgg	240
cggcacaccc	aggaacacca	cctatgaggc	ggaggcaggc	gactgc	acgaggacat	300
gaagaagtgc	gaggcgatca	tcccgtccca	ggccgggtgg	aacaaggcg	actcgacaa	360
gttgatgacc	aacttcgca	cgcggat	caccacc	aaggtaaaag	tggagaattt	420
ggcggaaatt	ccggaggaca	ttttgtcgaa	aggacacgc	gagggtgc	tgcagtccgg	480
taccaacccg	ttcgcgtccc	agaagggtt	cgtcg	ggtacccgac	gtgacgtgt	540
ccgtgggggg	gtgaacgt	acgtgtcg	gggcgactt	gagccgctt	cgaggaa	597

<210> 57

<211> 80

<212> DNA

<213> Globodera rostochiensis

AKK110P1

<400> 57
 ggcattgtgc gtctgcaagc cggtaacgaac aagttcgact cgcagaaggg catgaccctt 60
 ttcggtaacgg gccccgtcgtg 80

<210> 58
 <211> 513
 <212> DNA
 <213> *Globodera rostochiensis*

<400> 58
 gaattcgcca caccgctcac atcgcgtgca aattcgccga acttaaagag aagggtggacc 60
 gncggctcgg caagaaaattt gaggacaacc cgaagtcgtt gaagactggc gacgcccggaa 120
 ttgtcgaact gattccgacc aagccgatgt gtgtggaggc attcaactgac tacgcaccgc 180
 tcggccgttt tgctgttcgc gacatgaggc anactgttgc cgtggcgcg atcaaattcag 240
 tggagaagac ggaaggccgt ggcaaaagtga ccaagccagc gcagaaggtc ggcgcgactg 300
 gtggcgggaa gaagacatga ccaaggggag gggcggttcc ctaagggcca accgtcgacg 360
 aaaatcgac caaccttctt tttatcggtt tcttattttag ttcccttccac cggctcttat 420
 ccatattgtc ttgcgttgg ataatgtttt atttttttagt attgtccctgg ttggaaaata 480
 aatttggta attaaaaaaaaa aactcgtgccc gaa 513

<210> 59
 <211> 393
 <212> DNA
 <213> *Globodera rostochiensis*

<400> 59
 gaattcgttt gagcgaaaaaa aacataactat acaatggcaa caactgagaa gcctcagggt 60
 gttcaacagc cggcgtcagggt ctttggccga aagaagacag caacagccgt tgcgtttgca 120
 aaaaggggca agggctttagt caaggctcaat gggcgccctt tggactacat gcagccggag 180
 attctgcga ttaagcttcca ggagccaaattt ctcattgtt ggaaggacaa atttgaggga 240
 atcgacatac gaatccgcgt caaggccgtt ggacacatgt cgcaattttt tgcaattcgc 300
 caagcactgg ccaaggcact ggtcgcttcc taccagaaga atgtcgacga gcagagcaaa 360
 aaggaaactga aggagcaatt tggcttac gac 393

<210> 60
 <211> 154
 <212> DNA
 <213> *Globodera rostochiensis*

<400> 60
 cacgagccaa agaaattcgg tggacccggg agctcgctt cgctaccaga atcgtaccgt 60
 taagaaataa tttttagat caaatgtttt gatgatgatc ctttggggggg ttgttataaa 120
 aaaaaattta taaaaaaaaa ccggccatac tgac 154

<210> 61
 <211> 666
 <212> DNA
 <213> *Globodera rostochiensis*

<400> 61
 gtattccaag tttgagcgat cagagttctt caatctatta tcaactgttt tccatcaacc 60
 aactgtcatc atgcaaaattt tcgtcaagac gtcaccggc aagaccatca ctctcgagggt 120
 cgaggctagc gataccatcg agaacgtgaa agccaagatc caggacaagg agggcattcc 180
 gcctgtatcg cagcgtctga tcttcgcccgg aaaacagctt gaagacggac gcaccttggc 240
 cgactacaac atccagaagg agtccactct ccatctcggt ctgcgtctcc gtggcggaaat 300
 gcaaattttc gtcaagacgc tcaccggcaa gaccatcaat ttggaggtcg aggccagcga 360
 caccatcgag aacgtgaaagg ccaagatcca ggacaaggag ggcattccgc ctgatcagca 420
 gcgtctgatc ttcgcccggaa aacagctcga agacggggcgc actctggccg actacaacat 480
 ccagaaggag tccactctcc atctcgatc ttgcgttccgt ggaggagaga actgaatcgc 540
 gggctgatgg aaagatgacg aatatgtatgtt ctattcgatg acttgcgttct ttgcgtatataa 600
 ttgattgtgt tccatttgc ggtcatcaaa tctttatgac ccccttattt ggcattggaaac 660
 gataaa 666

AKK110P1

<210> 62
<211> 213
<212> DNA

<213> *Globodera rostochiensis*

<400> 62

gaattcgttt gagaaaacttt ttcaaccatt cattcaaatg tctcatcaag tgacacgggc 60
agcactcaac cacgggacgc gtgtactgag cgtgttggag aaggtaagt tggtctgctg 120
gtttgaggag acacattcgt tcgcgcaagt ggctcgaaga taccgggcag aatttggat 180
ggaaccaccg cagttggacc aagtgaagaa gtt 213

<210> 63

<211> 488

<212> DNA

<213> *Globodera rostochiensis*

<400> 63

agcaccggct caatcctcaa tggcacaacg acggcattct ccggcatagg agacggagtc 60
ggcttggag aacaacagcc aattcccgct gtaagcgatg cgggactgga tgcggaaaga 120
cagctgagaa tggccagaat gtgacccggg ggacctgaaat atttatgaac gaaattttcc 180
agtgaagtgg accaacgctc ttgcacttta tctgcttgc gtaaagtgtt tagaattcggc 240
ttccaattca aaggctttt attcccaac ttttattttt ggcacaaaaa tttcttagga 300
taagcgtgaa taatttattt attttttttt tcttctttt atctccgcct cgaagtcgca 360
agtgtccctt ttggcccggtt cccttttgc ttgaatgtt ttccattccc atcccctcac 420
tttctcatat ttgtgacatt cagctgcatt gttcgactcc cattaaaag ttgagtgaaa 480
tgcatttg 488

<210> 64

<211> 249

<212> DNA

<213> *Globodera rostochiensis*

<400> 64

wccrgakbng aacahcdkg vhwatnvcbn gschvbwagc rntgcsvddb wgnhnsswtg 60
gkqdryrbwnt msnwrmanrg artssstsgaa ttcccaagtt tgagagtaaa tattattagc 120
taaaaatggc agtcggaaaag aataagagaa tggcaaaaaa gggagccaag aagaaggctg 180
tcgatccgtt cacacgcaaa gaatggtacg acatcaaagc gcccggcgtt ttcacacatc 240
gaaatssts 249

<210> 65

<211> 362

<212> DNA

<213> *Globodera rostochiensis*

<400> 65

wcbrhdyb ytsgcrsnck tbdsbhcysy gcdwkmtnvk hscngdckty nyykkkbmr 60
ntmsnwrman rgartsstsg tcaaccgtac tcagggaaacg cgcatttcga ggcactttct 120
aaaaggccgc gtttacgaaat gtttactggg tgaccttaac agcactgacg ccgactttcg 180
aaagtccgc ctgatctgt aagaggtaca gggcaagatt tgcctgacca actttcacgg 240
aatgtcggtt actcgggaca aactgtgctc tattgtcaag aagtggcaca cgctcattga 300
ggcgaatgtg gcagtgaaga ctaccgacgg tttcatgctc cgactctttt gtatcggtss 360
ts 362

<210> 66

<211> 128

<212> DNA

<213> *Globodera rostochiensis*

<400> 66

aatcaaatta agaagacgag ctatgcaaaa gcctctcagg tgcggatgat tcgtgccaaa 60
atggtggaga tcatgcagaa agaggtctct tccggcgtat ttgaangaaa gtagtcaaca 120
agcctgtat 128

AKK110P1

<210> 67
 <211> 502
 <212> DNA
 <213> *Globodera rostochiensis*

 <400> 67
 gaattccatt aaaaaactaa acgaacaaat ctaaagatgg ccaccgaagt ggaggaaaaat 60
 gttcctacgg ttgaccatg ggggtctgtg gaggaagtgg gtggtaaga gtcgatgcag 120
 ttggtcagcc ttgacgttac cgaggtaaaa ctgttcggaa aatggtccct taacgatgt 180
 gaagtgtccg acatttcgct tggattat attgcggta agggaaaggc ggccaaatat 240
 ctgcccaca gcgcgcgcg ttaccaacag aagcgcttcc gcaaggccac ctgtccgtg 300
 gtggAACGGT tggcttgc aatgatgtg cacggggcga acaacggaaa gaaactaatg 360
 gcgggtgcgcg ttgtgaaaca ccccttcgag atcatcacct gctaccggag agaaccagg 420
 ccaagtgttgcgtg tgataaaacag tggggccnc gaagattnca cacgtatccg 480
 acgtgcggc actgttcgtc ga 502

<210> 68
 <211> 519
 <212> DNA
 <213> *Meloidogyne incognita*

<400> 68
 gcaaactttt atcaaataaa aaatttatat ttgccaaaca aatttatgaa taaaaattca 60
 ttaatcatta aaactacatt taaaatatac ttttagaga atgtcgtcta aaatattctt 120
 ttctccctt tatgcattcta tctaaccaga cttggaagca atatggctaa tcaagtcaac 180
 aatacggcag gaatacccaa actcggttac atacagactt accaatttaa caaaatgcgg 240
 gttgagaacc ataagagcct cggcgtcgaa aatagacgaa tgagtgtcgc caagaaagtgc 300
 ggtgaaaca acctggcct cagtatatcc aagaatccct ttaagctttc cttccgaagc 360
 agtcttaatt gcattctaa tagcctcctt cttgtgtcgc ttctccaaac gaggactcaa 420
 atcaacaacg aaaacgtttg ggcgtcgca caccggaaacg catttccgt aagcttccca 480
 tccaattcat ggattgaccc ttccaacagc ctttgcagc 519

<210> 69
 <211> 218
 <212> DNA
 <213> *Meloidogyne incognita*

<400> 69
 ttgattcttt attagtggac aatgacggaa gaccagaaga agttgcccgt ggtgcctgag 60
 actgttttga agcgaaggaa agttagggtc gtcagcgtg cttctctact caagaataaa 120
 ttggagaata ttaagaaggc taagttaaa acgcaagttt tctttaaacg tgctgagcaa 180
 tacttgattt catatcgacg taagcaaaag caagagtt 218

<210> 70
 <211> 293
 <212> DNA
 <213> *Meloidogyne incognita*

<400> 70
 taagaaagca gggaaattttt atgtcccaga tgaacctaaa cttgttttg ttgtgcgtat 60
 taaggaaatc aacaaggta atttaattt gctataaagt ttaggtggg ttttagacaat 120
 tcttccttt taatgcttc taacttttc aaaaaagttt tgattttac acccattaaat 180
 ctacaattt ttaatttcat cagatccatc ctcgtccctg aaaagttttt caactttttcc 240
 gcttgcgtca aatcaacaat ggagtttca ttaaatttga taaagctacatc 293

<210> 71
 <211> 422
 <212> DNA
 <213> *Meloidogyne incognita*

<400> 71
 aatgcaatta agactgcttc ggaaggaaag cttaaaggga ttcttgata tactgaggac 60
 caggttgcgtt ctaccgactt tcttggcgcactt cttttttat ttttcgtct tatgggtctc 120
 taagtttgcgtt ttttctaaat ttatattttaa cttttttat ttttcgtct tatgggtctc 180

AKK110P1
 aacccgcatt ttgttaaatt ggttagctgg tatgataacg agtttggta ttcctgccgt 240
 atttttact tgattagcca tattgctcc aagtctggtt agatagatgc ataaaaggga 300
 gaaaagaata ttttagacga cattctcaa aaagtatatt ttaaatgttag ttttaatgtat 360
 taatgaattt ttattcataa atttgggg 420
 tg caaatataaa ttttttattt gataaaaagg 422

<210> 72
 <211> 374
 <212> DNA
 <213> Meloidogyne incognita

<400> 72
 atctgagcat aaggaaactt ggccctcaagc tatagagcag accgattatg tggcaccgac 60
 tgagccaggtaa aactggact tcaacgttcc gcttattatg gattggctg ctgcttctga 120
 gtggcctcaa gaagaggaag cttaggtgc acctactgca ccaattggtc agccacagcc 180
 tcaacagcag caaactcaac aaggaggtga ttggaaactct ggtactatgt gatggtaag 240
 ggcaggaaaaa ttgatagaaa gagaattat tatggaataa atgtaatcaa ttttttttttgc 300
 tgattttttt gttacatata caacaagttt tttttttttt tttatataat aaaagttgtt 360
 aattaaaaaaa aaaa 374

<210> 73
 <211> 120
 <212> DNA
 <213> Meloidogyne incognita

<400> 73
 tttttttttt ttttttttca tcaatattttt gaagtgaaga accagaagta gttgcattcg 60
 agctttcaaa tttttttttt tgattactct ttaaacaaga ttcaactgat ggatctactg 120

<210> 74
 <211> 369
 <212> DNA
 <213> Meloidogyne incognita

<400> 74
 gtctaaccaa tcttagagcta ttccgttgcg ctgtctgttg attatttagat gttgattgaa 60
 cagcactagt ctctgatgtt gtttttttca atctcatttt taatgtatgtt agaggaagg 120
 tagaattctg attgtctatcg tcttttttttctt cttttttttaa tggcttttttca aatttatctt 180
 ctttttttttctt ttgtccat ttttttttcat ttttttttcaaa aggctcagga aatttttaattt 240
 cagacccgcgt ctttttttact gctgtatcta aaaaaaaccctt tcttaggcaac gtcccgatgc 300
 cactcaaattt caatttttttttgc cagatctaag tccttcttcc ttttgaacgaa 360
 attgaactg 369

<210> 75
 <211> 529
 <212> DNA
 <213> Meloidogyne incognita

<400> 75
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 aatctaaata aggctctatt ctaagtttat attttttttt tacataaaacc gtcaacccctc 120
 caagtttttca aatgttttgc ggtttttatg gatccctctgg taataatttg taggcttagaa 180
 aaaatgttgc agcaaaaagg aaaagcatca ttcttgctta ggcttctcca gcacattgcc 240
 tttcccccac accaaaaagctt attagctgtt cagttttttt taattttccct tcattgtcta 300
 tataacgttc agggtcaaaa tttttgggat ttgggtatata ttttttttgc aaaaagacat 360
 ccgataacttg ggggtatcata aatgtacattt taggcaacac aaactttcca acattcaat 420
 cttccaaggc taaaatgcctt aaattgaaaag ggactaaattt aacgagtctt aatgtttcat 480
 taacaacagc atttgtataa attaatttttgc caaaactaat 529

<210> 76
 <211> 449
 <212> DNA
 <213> Meloidogyne incognita

AKK110P1

<400> 76

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tcaacaatt	acttgacgca	gaaaagcgtg	ctgcagaaaa	gattaatgag	gcacgtaaaa	120	
gaaaggcaca	acgactttaaa	caagcaaaac	aggaagcgc	agctgaaaatt	gacaaatata	180	
gagaggaacg	tgaaaaaactg	tttaaagagt	ttgaacataa	ttacctcg	gctagagatg	240	
atattgctgc	aaaataaaag	cgtgaaaactg	atgagacgt	taatgaaaatg	actcgtagtg	300	
ttgtctctaa	taaacagcag	gtattgttc	gtctacttca	acttgtctgt	gacattcg	360	
cagaactgca	tcacaattta	caacttcaac	ttaagcttaa	tgaaaagcct	gcctaatttg	420	
tagttgattg	attataaaaa	tgaaattga				449	

<210> 77

<211> 643

<212> DNA

<213> Meloidogyne incognita

<400> 77

atttatattt	gaacaataaa	tttaacaaaa	aagtatggct	cgaggacc	agaagcattt	60
gaagcgtttg	gcccgtccaa	agaattggat	tttggacaaa	ttgggtggag	ttttgcccc	120
acgtccatg	tgcgggcctc	acaagcttcg	tgaatcgctt	ccttattt	tgtttcttcg	180
taatcgctt	aaatatgcac	aatcttataa	tgaagctagg	atgatttgca	aacaacgtct	240
cattaaagg	gatggcaagg	tgcgtacaga	aatgcgtttt	ccagctggat	ttatggatgt	300
ggtttccatt	gagaaaaactg	gcaaggtctt	tcgtcttc	tatgatgtca	aaggacgtt	360
cattactcat	cgcatacaaa	aggaagaagg	tcagcttaaa	ttgtcaagg	tagtaaagca	420
agcgattggg	ccaaaacaag	ttcccttat	tgttactcat	gatgcccgt	ctattcgct	480
tccggatcca	cacatcaagg	ttgacgacac	tgttgcgtt	gatataaaca	ctggaaaggt	540
tacagatcac	attagattt	ttctggtaa	tgttgcgtt	attactggtg	gtcacaaacat	600
gggacgtgtt	ggtattggtt	gacatcg	acgcccaccc	ggt		643

<210> 78

<211> 584

<212> DNA

<213> Meloidogyne incognita

<400> 78

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aaataataaa	ttggaaatata	ataaaaatga	aatttgagagg	caaaaagagc	aattaattcg	180
agatttgatt	gcctccttaa	cacgtgaaag	gcaatattca	cgagattggc	aacaatcaca	240
acagcaacaa	aatttcattt	acagtttgg	cccttccca	cattattcc	cctcttcagg	300
cattgaatgg	cccccaacaac	acaaaaaaaat	atttttggaa	gaaggggaag	tagaagaacc	360
tttagaggaa	aatgagaagg	aaaaaaagagc	acaaaacttt	gttcgtttc	gaaagagagc	420
acaaacattt	gttcggttt	gaaaaagggg	acagactttt	gttcgatttt	ggagagatc	480
aaaacatcaa	cataacttgt	cagatcagaa	gcagttaaaa	actgacaaac	aataaaaaatg	540
atgaattatt	taaaaatttt	tttaatgatc	tttaatttaa	aatt		584

<210> 79

<211> 556

<212> DNA

<213> Meloidogyne incognita

<400> 79

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tcccgcttga	tcaacacgt	ttgatcttt	ctggtaagca	acttgaagat	ggacgaacct	180
tggctgatta	taacatccaa	aaggagtcta	cacttcactt	agtttacgt	tttcgtgggt	240
gaaagggttca	cggttcattt	gctcgtctg	gaaagggtcg	tgctcaaact	cctaaggct	300
aaaagcagga	acataagaaa	aagaagcgcg	gccgtgctt	ccgtcgcatt	caatataacc	360
gtcgcttcac	caatgttgct	acttctgggg	cgggacgc	tcgtggccct	aactccaaac	420
ctgcataaga	gaatggctgt	atctgtatga	atgtatggtg	atataatcaa	ttaataacat	480
tcgactntat	gaagttttct	gttattcaag	ataaatctt	ttgttgaaaa	aaaaaccaag	540
tttgagatca	gttact					556

<210> 80

AKK110P1

<211> 424

<212> DNA

<213> *Meloidogyne incognita*

<400> 80

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aacattgttt taattaaaat ttaccccctcc ttagtcaatg acatcagaca gacttggccc 60
atgttcca gatttgacag cccaaagagac caacagactt gaacgaacta gttctttgg 120
cgatttggca attcgggatg gagttccata tccacctagg cctgcaatta ataatgttcc 180
tccatcacctg aatatgttga ctgcAACGTT ttctgtacca aatgtaaaatc agtacacggg 240
tgcataatgg tcttatacgac cagcaaatcc tggttataact tatatagtt ataaatgttca 300
ttttccgtat agaaattatc gaggtacac actgacggat gcttactggt acgaccgtt 360
tttattttt tcgccaat acaaacggtc aatgttccca attagattcc ggcattctga 420
ctac 424

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<210> 81

<211> 89

<212> DNA

<213> *Meloidogyne incognita*

<400> 81

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attatccaca cacctattgg agtaccctt accaaggaaa atggtacgac tatgacaatc 60
caacanatta cggccatcc tttgaccca 89

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<210> 82

<211> 168

<212> DNA

<213> *Meloidogyne incognita*

<400> 82

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tttttttttt taaaattttt tcattaacaa atgaccttaa cagataaaac ttaacagtca 60
aaagacaaca taatttccaa ctttttcaat attatccctt ttaacggttt gattttgcaa 120
ctcgctccaa ttgcgtccctc ttcttgatag catatgaatt gctcgaac 168

```

<210> 83

<211> 67

<212> DNA

<213> *Meloidogyne incognita*

<400> 83

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aattcatcg ccagacattc agcaattgtt ttgatattac ggaaagaagc ttcacgagac 60
ccagtac 67

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<210> 84

<211> 42

<212> DNA

<213> *Meloidogyne incognita*

<400> 84

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taacacgacg aagaggcgaa acatcaacag cctgacgacg aa

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42

<210> 85

<211> 429

<212> DNA

<213> *Meloidogyne incognita*

<400> 85

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tatacgagta gaatccccc gtggtcctcc attaataaca gcgcacaa gatattgaac 60
tggattctct ccagtcaaaa tatgtataat ttcaaaagcg tgcttcacaa tccgaacagc 120
catcaacttt ttaccatgt tacgtccatg catcatcatc gaacaaacca aacgttcaac 180
aatcggacaa tgagccttc gaaaacgttt gatttgatat cgaccacgac tgcgtccaa 240
atatttggcc gatttgctt taacagcaat ataatccact aaagaagcat cattaacttc 300
gatatcgctt aaagaccatt taccacaaat ttaatttca gaaaatcaa ttgttagtcat 360
ttgcatatcc cttgtccac caggaacatc agttgcggcc caattatcat cagcgggtaa 420

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AKK110P1

accatctcc

429

<210> 86
 <211> 435
 <212> DNA

<213> *Meloidogyne incognita*

<400> 86

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 aattttcttt tcatacattt ttaatttaaa aaacattta acaaattaca agaacaacaa 120
 acataattgt ctccctttt ttataaaaatt taaagttaa taagtttaa aacattctcg 180
 actggagtagc gtgtacttag tgtttttagaa aaggcaaaat tagttgtt gtttgaagag 240
 acaaattctt ttgcacaagt agcgagaaga tatcgagcag aatttggaaat ggaacccca 300
 catatggatt tagttaaaaa attacatcaa cgtttctca atactggttc tgtttctaat 360
 ggaataactg aacatttga agttaatcca acaatggaaa catcgacatc ctcaacagag 420
 ggttagcag atccg 435

<210> 87
 <211> 501
 <212> DNA

<213> *Meloidogyne incognita*

<400> 87

gttttttttt tttttttta aacaaaatat cgagtctta taagacaaaa ataaaagaca 60
 aaagcaattt agttttatca ataaaattaa aaatagtcaa tgtctcggtt cactcattag 120
 atttgtggcc ctaaagaggg ccgttgggt ttggttgtt tacttcagct gccttccacc 180
 aattgttctt tagccaccaa atccgtaaag agtacgtctt tggcggttca acgcatagac 240
 gacgtccatg gctgtgacgg tctttctctt ggcgtgtac caataagttt ccgcgtcgcg 300
 gatcacattt tcaaggagaaga ctttcagaac acctcgagtc tcctctgaaa tgagcccgaa 360
 aatacgttt actccaccc gacgtgcca gtcgggatt gccggttgg tgataacctt 420
 gatgatataca cgcaagactt ttccgggttgcg ctttagcgctt ccctttccaa gtccctttcc 480
 gcctttact cgtccggaca t 501

<210> 88
 <211> 270
 <212> DNA

<213> *Meloidogyne incognita*

<400> 88

ggaagtgtgt ttaagataaa tggatgatta gaaataaaaaa tgaattgatt aaaaattacg 60
 tttagaataat aatggatata ataaaaataa attggatgat ttaataaaaaa aaaaaaaagag 120
 agaaatagtc tcgagttttt ttttttttt tttttaaaaa ttaacaattt atctcatttt 180
 cctctccat gaaaattaac aaaaagacga caacttaatc ccataattaa catcatttt 240
 aagcttcagt cggcatgctt cgaataatgt 270

<210> 89
 <211> 286
 <212> DNA

<213> *Meloidogyne incognita*

<400> 89

caagcggttc ccaactcaat gttgtgcca tgataactcgta gaacaccagt tctcgccaa 60
 atagaatagt actcaatctc actgcgtcta aggcttggag tattattcga aataataaca 120
 agtttagctt ttccagaacg aagagtctt aacgtctgt ttagcccaa acaataacttg 180
 cccgatttgg taaccatggc gagacgagca ttgatatttt ctgtggactt tttctgttt 240
 ccaacaacca ttgttaacgca aaattaaaaat ctctttttta acaaat 286

<210> 90
 <211> 391
 <212> DNA

<213> *Meloidogyne incognita*

<400> 90

AKK110P1
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 tagacttgaa cgaactagtt ctttgggtga ttttagcaatt cgggatggag ttccatatcc 120
 tccttaggcct gcaattaaca atttccctcc atacacctaat atgttgactc gaacatttc 180
 tgtaccaaataat gttaaatcagt acacgggtgc aataggttct tatcgaccag taaatcctgt 240
 ctatacttat tatacgatata aatgctatTTT tccgtataga aactatcgag gctacacatt 300
 gacggatgct tattggtagc accgttatta ttatTTTcgt cctatataca aacggtaat 360
 gtttccaatt agattccggc actctgacta c 391

<210> 91

<211> 131

<212> DNA

<213> Meloidogyne incognita

<400> 91

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 caacaaatttta ccgcccgttc ttcgaccac gcatcagcgc atcattttca agacctttagt 120
 attacacatc a 131

<210> 92

<211> 571

<212> DNA

<213> Meloidogyne incognita

<400> 92

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 cctcaaaaaaa ttcattttgc gacgaccagc agcagggtgt tgctgctgtt gttgaccacc 180
 accccccttgc gcttgacctt gctgttgcgt tcccttcacg tcaacaggca aattgagttg 240
 caaataatca accatctctt tagtctctt atcaacacta atagttggat gttgagaagc 300
 atcaagatag gaaacttctg gaacccaattt atcacgacgc tcacgctctt cttcttgcaa 360
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 accagcaatt tgattacgca tccgtttgcgt aggaataaca gcaatttccct cacaatttcg 480
 ttgttccaca tgaaaatcat aagtcaagcg tgtataatat ttgtcaataa taacacgaga 540
 tgctttcttg acagtttga gagaaccgat t 571

<210> 93

<211> 671

<212> DNA

<213> Meloidogyne incognita

<400> 93

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 aattgcaaaag ggtgatgttt ttggaaaagga aacgcccattt gttctggtaa tggggatata 180
 tcctccaatg gccgaagtgc ttaaaaggagt ggaacttggaa ctttacgattt gtgccttggc 240
 gaatcttata gctgtcagac cagtcacgac tgaagaggca gcgttcaaaag acattgattta 300
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 tggtaagggtt ctgggttggta gaaatccagc aaatacaaaat gcttttattt gtgcaaaata 480
 cgcagcagat aaaattccag caaagaatgt cagcgctatg actcgcttgc accataaccg 540
 tgcatttgcc caaatagctg ctcgttgcgtt ggttgactgtt ggatctgtga agaaaagttat 600
 aatttgggaa aatcattcaa gtacccaaattt tcctgtatgtt aaacatgcta aagtaattaa 660
 aggtggcagc g 671

<210> 94

<211> 289

<212> DNA

<213> Meloidogyne incognita

<400> 94

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 agctgttattt atcgaaaaac gcaaaactgtc cagcgcaatg tcggcagcaa aggccggcatg 120
 tgcacattt catgattggc actttggaaat aaaagatggc gattgggtttt ctatggccgt 180
 tccttccatg ggttctttagt gaattccgga aggtttgatc ttctcatttc caattacaat 240

AKK110P1

tgatgcanaa acgcgtgact ggaaaattgt acaaagatta gaactcgat 289

<210> 95
 <211> 262
 <212> DNA
 <213> *Meloidogyne incognita*

<400> 95
 aatttaactt ttctaaacca aactttattt tttgtcttt atgtctactc aagtaccgat 60
 acgcgtgctg gttactggag cagctggta gattggttat tcttggta ttcaaattgc 120
 aaagggagat gtttcggga aagaaacgcc catcgctctg gtaatgttg atattccccc 180
 aatggccgaa gtgcttaaag gagtggact tgaactttac gattgtgcct tggcaaatct 240
 tatacgctgc gagccagtca cg 262

<210> 96
 <211> 323
 <212> DNA
 <213> *Meloidogyne incognita*

<400> 96
 aagacattga ctatgccttt cttgttggtg caatgcctcg aaaaagaagga atggaacgaa 60
 agatttact tgctgctaat gtaaaaaat ttaaatcgca aggactggct ctagcgaat 120
 attcaaagcc aactgttaag gttctggttt ttggaaatcc agcagataca aatgctttt 180
 tttgtgcaaa atatgcagca gaaaaaattc cgacaaagaa ttgcgtcgct atgactcgctc 240
 ttgaccataa ccgtgcattt gcccaaataatg ctgctcggt tttgggttgc tttgggtctg 300
 tcaagatagt tataatgtgg gga 323

<210> 97
 <211> 717
 <212> DNA
 <213> *Meloidogyne incognita*

<400> 97
 aatattttta acaaacgatg taacagaaaa acaaagtttt tttaacaaat ttcttgaac 60
 ctatttttt ttc当地acat ttttttattt aaattttaaac ctctcttcat ttctttaaa 120
 cacttcctg aactggaggt tcataagcat ctggacgact ttcaataact tctccacttg 180
 ctgtagttat agcaacttgt ccaccaccac ttccagcacc ctctccatgc atatccaaa 240
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 gaagtggata gaaataagaa caagactttt caatgtctt tccaaatagaa tcaggaattt 360
 atttgctgac aacttcctta agatcgcatg aagaacactc gcgatgata atctcaacca 420
 tccttagcagc aatttgacgc acttgagacg atttgcatc actagtctt ttcaacttggt 480
 ttggagcttt ctttgcataag ccaatacaga acaatcgaa gaaataacca tcagttttt 540
 tgacagcaac atttgcttca attaaagttt gccactttt gacaatagaa caaagcttgt 600
 ctcgagtaaa agtcattcca tggaaattgg tcaaacaac ttgccttga acctcttac 660
 aaataagtcg aaatttgcga aagtcaatcg tttttttt cgggtttt cagatcacca agagaaa 717

<210> 98
 <211> 758
 <212> DNA
 <213> *Meloidogyne incognita*

<400> 98
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 taacaacaaa caaaaatggg cgagcaagac aaaaagaaag ctggcggcg cgatgggtggc 120
 aaaaagaagg atggcttcga tgccaaaaag tttgcattt atttggctt tggaggaact 180
 gcccgtcgcc tttctaaagac ggctgtggcg cctatttgc tttttttt ggcgtggtaa tttggcttaac 360
 gttcaagacg cttctcagca catcgctgcc gataaactt ataaaggaat aattgtatgt 300
 ctgttgcgtg tgcccaaaga acagggagtc aactttgcgt tcaaggacac ttacaagagg 420
 gtgatccgtt actttccac gcaagctctc aactttgcgt tcaaggacac ttacaagagg 480
 atcttcatgg aagggtgtga caagaacaaa cagtttgc tttttt gatgtatgt 540
 ttatgatggca aaaattttct tgggtggaa agacacttacca gttgaagagt attttgcct 540
 ctgtgatagc tatacaacac tctttcaat tggagattca atgttgcgt gatgtatgt 600
 tagtaatctt ctgttacaat cacttaacaa cttcaatcaat tccaaatgc tggctcagaa 660
 ttataactcc tcaacaattt gcccgaagctt aaaaactacgt ttcaacatgc tacagctact 720

AKK110P1
tcaaaaatcga aacagattgt tttaaacgtt tgaaaattt

758

<210> 99
<211> 154
<212> DNA
<213> Meloidogyne incognita

<400> 99
ttgagttcgt tggcacattt gtttgttac aaaacgaaaa ttattggaa cgggtttcag 60
tgccatttct cgcaggattat tggcacttca cacattgtt ccaataacaa cgttaccgtt 120
tataatcaaa ctgttcctca aagttatgcc catt 154

<210> 100
<211> 125
<212> DNA
<213> Meloidogyne incognita

<400> 100
ttcagaatac tcaaggcttt atattcgttt ttgatagtaa cgacaaagag cgtattgtt 60
aagctcgtga ggaattgtat cgtatgttctt ctgaagacga acttcgcgtt tctgtactcc 120
tcgtat 125

<210> 101
<211> 219
<212> DNA
<213> Meloidogyne incognita

<400> 101
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aaatcgtaac tggtatatcc aggttacttg tgccacttca ggagatggtt tggatgaaagg 120
tttggactgg ttgagtaacc aattgaagaa tcaaggtaaa atgagtctaa ataaaaatgg 180
agaggggaaa gaggagaggt taattttta agaaaaaaa 219

<210> 102
<211> 473
<212> DNA
<213> Meloidogyne incognita

<400> 102
gtttttttttttt aattccaagt tttcttccaa atgagagaat agggagaatg 60
atggggaaa aaataggagc aagccaaaaa gccaaaaaaa aattttttt taaaatgatt 120
tttgtaaatg tggaaaagg tggatgtcaa ttgttagatc aaatgtcggtt gccttccttc 180
cactaaaatt tctctttctt ttctttctc ttctaaaatt cttccaaatgtt cgtccaaacg 240
aaatttcagc ctccctcttga tattccaact cccaaatacg cttccaaatgtt ttgcctttaa 300
cgtcacgagg agtaccaaatt ccagtcatca acttttgaga gtctcccttta ttccaacccg 360
cctgggatgg aattatcggtt tctgacttct tcataatcttc atatggaaatg tcgcccagact 420
ccgcctcgta tggatgttcc ctggcggtt caaaacctgtt catggccgtt tgc 473

<210> 103
<211> 114
<212> DNA
<213> Meloidogyne incognita

<400> 103
ttggaccgtt aggattgtcg ccaaagaaaa ttggagaaga cattgcaaag gcaacacaag 60
actggaaagg cttaaaggatc acttgcaat tgactatccaa aaaccgaatt gcca 114

<210> 104
<211> 255
<212> DNA
<213> Meloidogyne incognita

AKK110P1

<400> 104
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 acgtaaaaca cagtgaaaat ttgacgatcg agcaaattat caacattgcg cgccaaatgc 120
 gacctcggtc aatggcgaaa aaaattggaa gggactgtt agggaaattct tggcactgca 180
 caatctgttg ggtgtactgt tgatggacaa catccacatg atattgttga tgcaatccga 240
 agtggggaaaa ttgaa 255

<210> 105

<211> 571

<212> DNA

<213> Meloidogyne incognita

<400> 105
 tttttttttt tttttttttt tgcacaat aaatttactc agaaaaatca ttacaattt 60
 taacacacat ttttaattcc ttaatactcc aaaaaacttc tcttctttat tccctcttat 120
 tctcccaatt catttaaagt ttcatgtttg tgcggcgcc atgacgacgt tttgcattat 180
 agcgatatacg actgcccattt ttcatcgaa cccattgcgg cagcggcgtca ttttggtag 240
 cagccttagc cagcttgcgc ttgataataa acgtttgtg tgcagccatt aaattgttga 300
 ctttatccaa aattgtttt ttgaggcaaa taaacaattt taattttctt gctcaacaag 360
 tccatagcag ctcatctgtt caacaatctc cctcatgcgc ctcagtcctt agcgcttctt 420
 cttatgaatg tcaaaaacag cagaacaac ccccagcaga acctgttggc cttcttttgg 480
 aagttcatca atctggtcat tcaacaacaa cccttccatc tccatgtntc ttattacccc 540
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<210> 106

<211> 235

<212> DNA

<213> Meloidogyne incognita

<400> 106

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 agatagtgtat ggacttgata tggctaaaag tattttaaat tgaataaaagg aaaaagaagc 180
 attttaaaga aaatttagatg gaaatgctga agaaagaaaa aaatttttttta ttttt 235

<210> 107

<211> 702

<212> DNA

<213> Meloidogyne incognita

<400> 107

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 tttgaaaaaa aaaaaaaaaa aaaaaaaaaac tcgagaagaa atccttgccg aaattgacgg 120
 ctctcaaattt gaggagtatc aacgtttctt cgatatgttt gaccgtggaa agaatggcta 180
 tattatggct actcaaattt gggtaattat gaatgctatg gaacaagatt ttgatgaaaa 240
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 cgaatttcgc gctttgtt acactgttgc gaataactgtt gacaaggaca ctttgcggaa 360
 agaattgaga gaagcttttgc tcttttgc aaaaaggggc aatgttaca tttctcgatcc 420
 aacactcaaa ggattacttc acgaatatcgccc cccagaccc agcgataaaag acttggatgc 480
 cgcagtagac gagatcgacg aagacggaa cggaaaaattt gaatttgaag aattttggga 540
 gttaatggct ggagagactg attgaaattt taatttgaat gactagaaaa ttgactaaaa 600
 tattttgcca ttaaattttt gaaagtgcctt aaaaattgcct ttcttgaaat ttttattttt 660
 aacgtctaaa taatgataaa aatggatata aaaaaaaaaaa aa 702

<210> 108

<211> 423

<212> DNA

<213> Meloidogyne incognita

<400> 108

aaaattaaaaaaa taaaagacaa acaaataaaat ataaattaaa taaaataat taaaataaaac 60
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 agaaagaaaaa tgccaaaggg gatgagaac ttgttgaaga aaaaagttca aaaaatataaa 180
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ccccatttgt	tgttgtagtc	catggagaga	aagcaatttc	cccattcga	aatgttgaac	300
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aattatgagg	tttgttgttgc	tcctgacgtt	tttattgtc	tggagctggg	tgaggatcac	420
caa						423

<210> 109

<211> 994

<212> DNA

<213> Meloidogyne incognita

<400> 109

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gtcaaaaact	gccaaggccg	aattcaagcc	tttaaaacgt	tcaagagaag	agcaaaaaaga	180
tggaaattgaa	cttgcgtcgt	catcgtaaa	ggggaaaattt	attattaaag	caaacaaaaaa	240
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ggaagaggta	gatgaactaa	aattggatga	ttggcgttgaa	aatgtgcga	agataataac	420
gaaattcaga	taaaaataac	aaagaaaatgt	ttataaaataa	agctgagttt	gccgatatcg	480
accaaaaat	tttgtatctt	tttacagaaa	tttgtcaagt	ttttaagaaa	tatagaagt	540
gacgtattcc	aaagacgttt	aaatgttattc	caactttgtt	tgatggggag	aaaattatcg	600
aattaactcg	cccagatgt	ttgtcggcag	ctgcaatgtt	acatgttacc	aaaatattt	660
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ttcgagatga	tattgacgga	ttaaaaattt	acatttccat	atgtatcaat	gcttattttaa	780
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caatttttct	tttcgagaag	ctgttgttct	tgcttctatg	cttcgtaaag	cctccatccc	900
tcaatttacac	gcggccgcag	catgttgtgag	tatttttgtt	tttagaatata	cttcttcaag	960
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<210> 110

<211> 476

<212> DNA

<213> Meloidogyne incognita

<400> 110

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aaattgagga	gtatcaacgt	tttttcgata	tttttgacgg	tggaaagaat	ggcttatatta	180
tggccactca	aattggggta	attatgtatg	ctatggaaac	agattttgc	aaaaaaactc	240
ttcggaaaattt	aatccggaaa	ttcgacgcag	acggcagcgg	caaaatcgaa	ttcgacgaa	300
tctgcgcctt	ggtatacact	gtggcgaata	ctgtagataa	ggacactttt	cggaaaagaat	360
tgagagaagc	ttttcgcttc	ttcgacaagg	agggtatgg	ttacatctct	cgtccaacac	420
tcaaaggatt	actccacgaa	atcgccccag	acctcagcga	taaagacttg	gatgcc	476

<210> 111

<211> 189

<212> DNA

<213> Meloidogyne incognita

<400> 111

cgaagacgga	agcggaaaaaa	ttgaatttga	agaattttgg	gaattaatgg	ctggagagac	60
tgattgaaat	tttaatttga	gatgaaataaa	aaatattact	aaatattttt	ccataaaaatt	120
ttggaaatgt	ccaaaaatttgc	cctttttgag	aatttttattt	tttaacgtct	aaataatgaa	180
taaatggat						189

<210> 112

<211> 164

<212> DNA

<213> Meloidogyne incognita

<400> 112

ttgaggaaat	ttaatttttt	aaacaaatat	aataattacc	aaacaacaaa	aaagaatccc	60
aaaaacaaca	tttttaaaatc	aaatgacaga	catatatttgc	caataacgt	gtgtggattt	120
tctttttttt	taaataatttta	acatctttag	cctgtatctt	cttc		164

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<210> 113
<211> 539
<212> DNA
<213> *Meloidogyne incognita*

<400> 113	cagtttctg	cgcagatttgc	gtaacctttc	caccagcttc	gaccttctcg	acggccttga	60
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	agtcaaaa	agctcaaca	cacattggct	tggtggaat	taagtcaca	ataccacat	180
	ctccagtc	caaagcctt	ggattgtctt	caacccctt	tccagttcga	cggtcgac	240
	tctctttaa	ctcagcgaac	ttgcaagcaa	tgtgagcagt	gtgacagtc	agaacaggcg	300
	tgtagccagc	agcaatctgc	ccaggatgtt	tcatgtat	aacctgaga	gtgaattgt	360
	tggtctcc	ttctgggtca	ttcatagat	cagaatgtac	tgaaccacgt	cggatgtct	420
	tgacagagat	gttcttaacg	ttaaatccaa	cattgtcttc	aggaacacgt	tcagggagag	480
	actcgtgtgt	catctcaaca	gatttaactt	cagtagaaaat	tccttcagga	gcaaaggta	539

<210> 114
<211> 314
<212> DNA
<213> *Meloidoqyne incognita*

<400> 114	gttttaatt	ttagaaaatg	tctacagaáá	cagaaaagga	tttagaacgt	tgggaggatg	60
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	cggagaatct	tgaaaagacta	aggaaaatgtc	cagtttgggt	tgttggtgtct	ggtgngcttg	180
	gatgtgaat	tttggaaaaat	ttggcccttat	caggatttca	aaatattgaa	gttatgtata	240
	tggacacaat	tgaccttca	aatctcaaca	gacagttttt	gtttcgtgaa	cacgatgttg	300
	qcttatacaa	aqca					314

<210> 115
<211> 200
<212> DNA
<213> *Meloidogyne incognita*

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<400> 115
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gacttgcatt ttatggggca ttttcaatta taatttgggg actagattct attgtatgctc 120
gaagatgtt aaacgcgccaca gggtgttctt tggtcgaatt tgacgaagaa aacaagccac 180
ggccggccac aatttttcca                                         200

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<210> 116
<211> 471
<212> DNA
<213> *Meloidogyne incognita*

<400> 116	tttggtcgaa	aaaagactgc	tactgctgtg	gcatttcca	aaaagggaaa	aggattaatc	60
	aaggcaatg	gccgtccctt	agaatttttgc	caacctgaaa	ttcttcgtat	taagctacaa	120
	gagccattgt	tgattgttagg	aaaggacaaa	tttgcggaa	tggatattcg	catccgtgtc	180
	aaaggtgtgt	gtcatgttgc	acaaaatttat	gcaattcgac	agtcaattgc	taaagtttgc	240
	gtggcctatt	accggaaaaa	cgtggatgag	caaagcaaga	agaattgaa	ggatcaactt	300
	gttgcattatg	atcgtaatttgc	gttgcgttgc	gatccggac	gtcaggagcc	aaagaattgt	360
	ggaggacctg	gtgtcggtgc	tcgttatcag	aaatcttatac	gttagaagtt	atgaaatttat	420
	aaaattgtgt	gttacgaaatt	aatttttatttgc	tttgttggat	aaatntgaaat	a	471

<210> 117
<211> 593
<212> DNA
<213> *Meloidogyne incognita*

<400> 117
gaattcaaaa aatattaaaa ttgtttataa taatttctaa aatqaaqccca aaggttggaa 60

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tccaagttgt	ggctgtcaat	gaccgggtca	ttgatcttga	ctatatggtc	tatatgttta	180
actatgattc	caccacccgga	cgctttaaag	gaaagattca	agcaagcaat	ggaaatttgg	240
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aaaagattga	ctgggcagg	tctggtgcgt	attttgttat	tgagtcgact	ggagttttta	360
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ccaagcatca	tatcattagt	aatgcttcct	gcactactaa	ttgtcttgct	cctttgcga	540
agtttataaaa	tgacgagttt	ggcataatttgc	aaagttgaat	gactactgga	cac	593

<210> 118

<211> 576

<212> DNA

<213> Meloidogyne incognita

<400> 118

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tgagggggaa	gtaaaaatgaa	agaagggaga	gagatatgaa	ttggaggttt	ttttgttaaa	180
ataaaatttt	ttttctgtaa	aattcttccc	gtttctgagc	ttttcgtct	tttttcaatt	240
ttcgtttgtc	gaaactactaa	actttacaat	ttggtaggt	tctttttgtg	aaacataaaat	300
atctccatta	tcgctgattt	caagggcatg	ggcgttttcg	agacccttgg	caaagctatt	360
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attttcagcc	tttgtgaaga	aagtgcctgt	ggggacgtaa	gcacgtctat	tttgggtttg	540
agcgccttct	aatccagcag	aaaagcatttgc	aatacgttgc			576

<210> 119

<211> 559

<212> DNA

<213> Meloidogyne incognita

<400> 119

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tttttgggtt	tttcaacttgc	gtcaggcttc	ccaaatttgc	tgagcaattt	cccatcttgc	120
tcaaacttca	ttattctggct	attacagtaa	ccatctggca	cgaaaaactc	tcctgtactg	180
gcaatagcaa	cgtctgttgg	tttgcaaaaaa	tgtttgcatt	ctgtccctgg	aacaagctt	240
tcgccccaaac	tcataattaa	tttaaaatcc	ttgtcaagtt	tgtggacttg	atgacttcca	300
acgtcagtaa	cccaactatt	gccgtgggca	tcgatttttgc	gtccatgagg	catgtaaaac	360
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gtgttttggaaa	tgatgcccac	ggatctgttt	aggtggttgt	tctcatcaaa	cgaaaattca	480
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<210> 120

<211> 366

<212> DNA

<213> Meloidogyne incognita

<400> 120

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tagttgtgt	agcgggttttgc	gtgtctgtgt	tatgtatggat	caagcaagta	aatacaatgt	120
agttgaaggaa	gaactcccttc	ttaattggat	taagaaatgt	acaggcgaaa	atattgttat	180
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tgtcaacaaa	attgtccaa	attcaatcac	aaaggccacag	gcaaaaccga	acagcacatt	300
ccaatataatg	agcaatttgg	agctgttctt	aacattttat	tcaagccaag	gagtccctag	360
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<210> 121

<211> 661

<212> DNA

<213> Meloidogyne incognita

<400> 121

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ggtaCTggag	accaagtgcg	ccttcgtgtt	taaagatggg	aaattgaaag	aattttgggt	180
aaacataata	aaaagacatt	ttatggcaat	aaaaaaatgt	aaaaaaagct	tgtcttttaa	240
atattttggc	aaaacatTTT	actttcacaa	aattttaaa	taaattttatg	aagattgttc	300
cgtcacttC	atcattttccg	atcgaccttt	gttggTTCT	aagtgcTTG	gcCAAAGAAA	360
ggatatgtaa	aattgaattaa	tgaataaaaaa	taaatcactc	aatcagaggc	attgttagtc	420
tctcacttcc	tcctctttac	ccattggcta	accagctta	aggatTTTT	ccataagttc	480
aagggtgtacg	taaatcgaat	accgactgtg	gtatcttaat	tttccatga	aattctccaa	540
aaaaaaaaaa	ttttttttat	ttttttccaa	taatgctatc	tatattttt	gcttttaatc	600
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a						661

<210> 122

<211> 173

<212> DNA

<213> *Meloidogyne incognita*

<400> 122

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ggggaaattgc	tcagtcaatt	tgggaaggct	gactccagtg	aaacacccaa	aatggagaa	120
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<210> 123

<211> 584

<212> DNA

<213> *Meloidogyne incognita*

<400> 123

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gagaaaagaac	acttcctcgt	cggcgtcacc	aatcaagatc	agggcagtc	attagaatcc	180
caagttttcg	taatggatat	gaacacaggaa	agggtctaata	gcttgctaa	gggtctagaa	240
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ttgtgcacaa	aaaaatgggc	gggcggggcga	atggctggc	aaaggatggc	gataaaatctt	540
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<210> 124

<211> 650

<212> DNA

<213> *Meloidogyne incognita*

<400> 124

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ctcttctcaa	gaagtacttc	acgaaggaag	ttatggacca	gtgtaaaggg	ctaaaaacta	180
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agaattatac	ggaaatgcata	gacaaaggta	aagggttttt	tgagcagctt	aagtctgtatg	540
ctgagcttgg	tggcacctat	tatTTTTG	agggaaatgac	caaagaggtt	caaactcaat	600
tgatcaagga	tcacttcc	ttcaaagaag	gagaccgtt	tttgcaagct		650

<210> 125

<211> 1013

<212> DNA

<213> *Meloidogyne incognita*

<400> 125

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gttttgaatc	aaataattaa	attttaaatt	atttaaacag	ctacacgagg	cctcagcc	180
ccccgttgca	ttcaaattgg	tcggcacgg	tggcgtat	aattttattt	tttaggtaat	240
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accaaacatc	cggaatacaa	ccacgaagtt	aacattgacc	aaagcgaaat	tcctttgcaa	960
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<210> 126

<211> 80

<212> DNA

<213> Meloidogyne incognita

<400> 126

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cttaccaaat	gggatcaat					80

<210> 127

<211> 585

<212> DNA

<213> Meloidogyne incognita

<400> 127

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aagcgggcat	gacagggttt	ggaactccaa	ggaacacaac	atacgaggcg	gagictggcg	180
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ataagggaga	ctctcaaaag	ttgatgactg	gatttggat	tcctcgat	gttaaaggca	300
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aaaatttttt	ttggctttt	ggcttgc	tattttttcc	ccccatcatt	ctccctattc	540
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<210> 128

<211> 287

<212> DNA

<213> Meloidogyne incognita

<400> 128

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cgcgtttctg	gtactttat	agtagttgc	gtcgtgtt	gaagactaac	ggagagat	180
tttcgtgtca	ggaggtttt	gaaaagaaga	taggctctgt	aaagaattat	ggaattttgc	240
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<210> 129

<211> 175

<212> DNA

<213> Meloidogyne incognita

<400> 129

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AKK110P1
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 <213> Meloidogyne incognita

<400> 130
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 ggagcaatag aacgcttgcg tcggcgaggc tcctcagccc tagtaacgtg aaatttctt 180
 gcaatcatcg atttgtgtag tccatctttg gctaagacct gttctaagtgc ttgttcataat 240
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 tctcgactgg agtatccac agggcaggga ttggagggt cacaatatgc tggcaaaaca 420
 ttgtcaactct taatctcttgc gcggtgtgaa aattcagatt ctggatggag ttgttggtct 480
 ccttcaccgg cacccctgtcataaattta tgtccaaacg caatgggccc ggaagcactt 540
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<210> 131
 <211> 466
 <212> DNA
 <213> Meloidogyne incognita

<400> 131
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 tcctgttaggt ggttagggcgc ggcctgctgc gccatataatc attgggatca gcattgtcag 420
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<210> 132
 <211> 266
 <212> DNA
 <213> Meloidogyne incognita

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 tgcggctttt tgaatatggc ggtttccgc ctgaagcgaa ttattttatc ttgggtgatt 180
 atgtggatag aggaaagcag agcttggaga cgatttgcgtt gctgttgcc tacaagatca 240
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<210> 133
 <211> 308
 <212> DNA
 <213> Meloidogyne incognita

<400> 133
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 tggaaaacat ttactgttgc ttcaattgtt ctgccaatttgc ctgtgtatcgatgagaaa 120
 atatttgtt gccatggagg tttgtcaccat gatttgcaga atatggagca aattcgaaga 180
 attatgcac cgacggatgtt gccagataca ggtcttcctc ggcacccctt atggtctgtat 240
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<210> 134
 <211> 335
 <212> DNA
 <213> Meloidogyne incognita

AKK110P1

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 tttttaggg ttaatgact ggacttataat acaatcaacc aatcgatcct attcaattt 180
 tggagaatgc aatagctaaa cttcgaaaaa atcctgatct tccattaaag tggatactt 240
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 atcgatgttc ttataaacaag agactcccta tcgaa 335

<210> 135

<211> 506

<212> DNA

<213> Meloidogyne incognita

<400> 135
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 ttcctgagac tttgatcacc ttcaaaacat taaaacgaac agtttactc aaaggcctgc 180
 attcaccgat cgtgacaata tcaccaatag agatatcag gaaacatggc gaacagtgaa 240
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 cacgccaat gacaatttg cgctgcattt tggttcttgc aacaacacca gtcaaaaatac 360
 ggcacgaat tgaaacattt ccagtggaaag gacactttt gtcaattaa ttgccttcga 420
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 tcgctnttt gccaatccct tgcgtc 506

<210> 136

<211> 230

<212> DNA

<213> Meloidogyne incognita

<400> 136
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 tttttttgtt ttttaattgt ttattttgc tactaattttt cttttaattt gccccttacc 180
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<210> 137

<211> 216

<212> DNA

<213> Meloidogyne incognita

<400> 137
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 tcactcgctg tttgtaccaa ctctacttagc tgtaattcgt ctttagctgt gcccgttattt 120
 tcttagtgaat cggaaagaaag tgatgaacaa caaaagacgg gggatggac aaatctaaca 180
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<210> 138

<211> 395

<212> DNA

<213> Meloidogyne incognita

<400> 138
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 ctctgttctc gttatgtgtc acactcgca acttgcattt caaatttcaa aggaatata 180
 aagatttgc aatataatgc cggaaactaa gtttcgggtt ttctttgggt gtatgccat 240
 caagaaggac gaggagactt tggctaagaa cactccgcac attgttgcgt gcaactccagg 300
 gcgtctgctg gcgttgggac gtacaggaca attgaagctg aaaaacatca aattcttcgt 360
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<210> 139

<211> 591

AKK110P1

<212> DNA
<213> *Meloidogyne incognita*

<400> 139

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gactggcagt cacttactga agtcaaggaa atgggtctta tgctgttgaa ctgctcctgt 180
ttggcgtggg aactgagcaa aatatttgcg atttactggc ggattggaca gaatcacaat 240
cgcttgcccg ctgtttggcc agtttattta caatcaaaaat tcaacgctca acacccaaatg 300
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aacccaaagg gcagagaaca cgacctttcg gccatatgct catgcatggg aaaaggccaac 420
gaatttgttc gaattgcgggt aatggattat attcctgcaa caatttacat gccgaatgg 480
aacaacatata attggccatc gatcgatgac gcgataagaa cggcagctta tcgggggtgtg 540
aaagttgacc tttggtgagt ctgtggcccc atttgaatga acgagcgatt t 591

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